IDENTIFICATION OF SPECTRAL BANDS TO DETECT NITROGEN AND PHOSPHORUS DEFICIENCIES IN WINTER WHEAT

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IDENTIFICATION OF SPECTRAL BANDS TO

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ABSTRACT

Past research in winter wheat *(Triticum aestivum* L.) showed the potential of spectral indices to detect winter wheat phosphorus (P) status although no specific indices were developed. An experiment was conducted at Perkins, OK in 2007/2009 to identify single or combined spectral indices that can detect nitrogen (N) and phosphorus (P) as well as P independent of N deficiency in winter wheat. A randomized complete block design with three replications was employed. Treatments included twelve factorial combinations of three rates of P (0, 34 and 67 kg P ha^{-1}) and four rates of N (0, 56,112,168 kg N ha^{-1}). Four types of spectral radiance measurements were collected and these included a full bandwidth spectrometer (300nm to 1100nm), Greenseeker™ sensor, New-experimental 4 band sensor, and digital pictures at four different winter wheat growth stages. Forage and grain yield were collected and measured. Forage and grain N and P as well as postharvest soil residual P contents were determined. Correlation analysis was used to test the relationship between spectral readings vs. forage and grain yield, and forage and grain N and P content of winter wheat. Similarly, stepwise regression procedure was used to select wavelengths and rations of wavelengths that can detect winter wheat N and P status. Analysis of variance (ANOVA) was employed to test the effect of N and P rates on several variables. Spectral reflectances at certain wavelengths were identified from spectrometer data and indices that can detect N and P status of winter wheat were developed. Spectral Phosphorus Indices: SPI1 and SPI2 were

developed from 915 nm numerators to 455 nm denominators, and 865 nm numerators to 505 nm denominator wavelengths, each averaged from10 nm bandwidth, respectively were significantly correlated with winter wheat forage P status over the two-year study. Also, these indices were significant for forage N content. Reflectances at single wavelengths, each average from 10 nm band widths between 605 to 695 nm were detected forage P content at Feekes 10 in 2008 and Feekes 7 and 10 in 2009 while the reflectance at wavelengths from 455 to 715 nm and from 815 to 925 nm were consistently correlated with forage N content at the above mentioned growth stages. There was no index, except the promising result of picture index (R/G), that could detect winter wheat forage P content independent of forage N content using the above instruments in the two-year study. This was likely because 1) wavelengths that detect forage P content were found within the range of the wavelengths that can detect forage N status and, 2) Nitrogen rate affected crop biomass and resulted in forage P content dilution as the crop grows.

CHAPTER I

INTRODUCTION

New time and cost effective technologies are needed to solve the problem of crop nutrient deficiencies like N and P because deficiencies of crop nutrient especially macronutrients can result in reduced yield or sometimes total yield loss. To alleviate this problem, in-season or real time assessment of crop nutrient status is important. Real time crop nutrient status is predicted from crop canopy reflectance captured by sensitive devices. Those devices were first introduced into agriculture through satellite imagery to estimate crop acreage and yield.

The United States Department of Agriculture (USDA) started using satellite imagery in 1972 when the National Agricultural Statistics Service (NASS) applied it to improve statistical precision of crop acreage estimates in 10 states. Since the 1970s, ground sensors of the same bands that were used by USDA were designed and used to diagnose crop health (Milton, 1987). Gradual development of this technology increased the spatial, spectral, temporal, and radiometric resolutions to the level of *in-situ* measurement.

Real time sensing provides accurate information on crop health condition, spatial distribution, and expected yield. Of the common uses of remote sensing data for crop assessment, crop greenness is the most common, and involves the comparison of reflectance of the visible portion (red) and infrared portion (near infrared) of the spectrum (Wittich and Kraft, 2008).

To date, the research conducted on sensing plant forage P status is limited and its spectral response to specific or a combination of bands has not been determined. According to Deleon (1999) different wavelengths that were correlated with winter wheat forage P content were not consistent overall growth stages of the plant and in different years. Better consistent correlation between forage P content and spectral reflectance has been reported in the region of the near infrared (NIR, 705 to 725 nm numerator) and visible (505 to 515 nm denominator) spectra, and 430 nm for forage P content. Likewise, DeLeon (1999) showed that 755 nm was suitable for predicting grain yield. Also, Girma et al. (2005) found that the wavelengths 515/675, 555/675 and 805/815 were predicting 94% of the difference of cheat and raygrass from wheat crop at Feekes growth stage 3 while 805/675 and 755 were predicting 66.7% at Feekes 5 growth stage.

Research on winter wheat and bermudagrass (*Cynodon dactylon L.)* showed the potential of the spectral indices to detect plant P status although no specific indices were developed. Additionally, some environmental problems such as weed presence, and cloud cover were affected the results of the study. Furthermore, the authors suggested that sensor sensitivity might have contributed to their failing to identify specific indices (Sembiring et al., 1998).

Consistent reflectance at certain wavelengths that can detect winter wheat forage N and P either as separate or as interaction must be isolated. Alternative days and hours of the day during crop measurement is important to reduce environmental impacts such as rain, cloud cover, and crop shadow effect that can affect the result. Also, objects that can increase reflection to the sensitive sensor like flags should be removed during crop reading. The scope of this study was limited to searching indices that can detect winter wheat forage N and P content.

OBJECTIVE OF THE STUDY

The working hypotheses of this research were (1) The status of N and P in winter wheat can be detected by indices developed using a full band spectrometer (300nm – 1100 nm) spectral reading, and (2) Phosphorus deficiency can be detected independent of N deficiency using spectral indices.

The objectives of this study were (1) to identify wavelengths or combination of wavelengths that can detect midseason winter wheat N and P deficiency using a full band spectrometer (300nm – 1100 nm), and

(2) to identify spectral bands or combination of bands that can detect mid season winter wheat P deficiency independent of N deficiency.

CHAPTER II

REVIEW OF LITERATURE

Phosphorus and N are two of the most important nutrients required to sustain vital physiological processes in plants. Since P is a constituent of nucleic acids; it can influence cellular division and development particularly in grain crops. Also, it is the medium for cellular energy production, accumulation and transfer through ATP (adenosine triphosphate) in addition to photosynthesis (Beegle, 2007). Also, N is important in determining wheat crop tiller and kernel number, kernel size and yield (Franzen, 1997).

Plant P deficiency symptoms can be visually identifiable but difficult to identify at lower P rates (Rehm and Schmitt, 2002). The purple color of the lower leaf margins of the younger plant, retarded growth, and late maturity are the main indicators of P deficiency (Beegle, 2007). Recently, visual analysis of crop nutrient deficiencies have been replaced by indices developed from remotely sensed (sensing plant without physical contact with the device from specified distance) plant canopy reflectance, which shows better results for crop nitrogen status (Raun et al., 2002; Eitel and Long, 2007).

Sensing is a result of the interaction of light energy, target object (plant), and the sensor. The target object may have three or fewer characters based on the wavelength and the object quality; absorbing, reflecting, or transmitting the light energy incident up on it. In the case of plants, the above mentioned characteristics are common.

Plant biomass is the most important factor that determines the fate of the radiation that strikes the crop canopy. Sensors currently available to collect the reflected radiation are sensitive to this biomass and the chlorophyll that determines the color of the plant. Jones et al. (2007) noted that density of crop biomass and plant chlorophyll concentration affected the NDVI collected using a Greenseeker™ sensor and multispectral imaging system. Factors that affected biomass and chlorophyll of plant like freeze and drought also affected the correlation between N uptake and the normalized difference vegetation index (NDVI) (Stone et al., 1996a). Similar results were reported on the correlation between forage biomass and forage N content and spectral reflectance readings (Stone et al., 1996b).

At Feekes 4 and 5 winter wheat growth stages, the percent ground cover of the plant was above 50 and 60, respectively (Raun et al., 2001). As the ground cover gets sparse, the reflectance from the soil surface is what the sensor detects. Reflectance from the soil occurs at different wavelengths compared to a growing crop. The author also reported the importance of a closed canopy in decreasing soil reflectance and increasing the NDVI value in corn plant starting from V6 (unfolded 6^{th} leaf of corn) corn growth stage (Raun et al., 2005). In another study, it was reported that increasing crop biomass with growth stage increased the NDVI value from the sensor (Martin et al., 2007).

In healthy green plants, in the visible portion of the electromagnetic spectrum, chlorophyll absorbs much of the red light energy and green is reflected back (Thomas et al., 2004). This absorption of light energy differed based on plant growth stages. For instance, at younger growth stages, wheat had deep green chlorophyll. As wheat grows, at later growth stages the chlorophyll pigment decreases particularly on the older leaves and absorption of light in the red region of the visible spectrum decreased even in the healthy plants (Babar et al., 2006). The color of the crop was not only affecting spectral readings but also affects grain yield. Girma et al. (2006) reported that the color of the crop from mean leaf color and chlorophyll meter (SPAD) that has been collected during Feekes 5,7, and 10 wheat growth stages showed a positive correlation with the final grain yield more strongly with the color value at Feekes 7 ($r > 0.85$).

Li et al. (2004) reported that the reflectance in the visible region of the spectrum (350 to 704 nm) was negatively correlated with winter wheat ground cover while the relationship was positive above 730 nm with high correlation coefficient at 784 nm. Their results suggest that increasing crop biomass decreased red reflectance in the visible region while it increased the NIR reflectance in the infrared portion of the spectra.

For those plants under stress, the greenness is less when evaluated on the basis of normal or healthy vegetation. But the capacity of visual analysis of relative stress is very low when compared to scientific devices that have been developed for crop scanning purposes like radiometers and spectrometers, (Milton, 1987). Using these instruments, different indices such as NDVI, the most common index for crop and vegetation assessment, were developed.

Field spectrometry is an important tool in the field of remote sensing although it has methodological problems in field data collection. Field spectrometry has the potential to work in three areas of remote sensing, calibration, prediction and modeling (Milton, 1987). Before measurements were started with spectrometers and radiometers, information on different spectral signatures were developed from different representative features and/or materials in the laboratory to obtain a threshold on absorbance and reflectance. Spectrometer data collected in the field is under the influence of atmospheric effects and solar illumination (Nicholas, 2007).

An experiment on corn plant canopy reflectance and leaf greenness, using hand held multispectral radiometer readings in 11 visible (460, 507, 559, 613, 661, 706 nm) and IR (769, 813, 850, 900, 950 nm) bands showed a positive correlation with leaf chlorophyll measurement (SPAD) and canopy reflectance (NDVI) with crop nitrogen status at v6 for early nitrogen application demand (Ma et al., 2005). Kruse et al. (2006) studied bent grass forage N content using spectral reflectance and statistical models and they found that the use of spectral analysis like NDVI was not reliable because of lack of consistent measurement over years. However, they suggested that advanced remote sensing systems involving canopy reflectance have the capacity to accurately distinguish crop nutrient stress and simplify the prediction to correct the required nutrient.

For assessing crop N status, green (550 nm) and red (675 nm) were the most important wavelengths from the visible portion of the electromagnetic spectrum while NIR (780 to 810 nm) was important to determine amino acid ($R\text{-}NH_2$) content or concentration. Also, for winter wheat, red (660 nm) and NIR (780 nm) wavelengths had good correlation with total N up-take (Stone et al., 1997). However, healthy vegetation reflects most of the NIR radiation and absorbs most of the light in the red band for photosynthesis. Thus, the proportion of the amount of red absorbed and NIR reflected determines crop health (Thomas et al., 2004). Osborne et al. (2002) studied the wavelengths in the visible (red and green) and the NIR bands and then categorized them in two different indices; N content and yield estimation respectively. The researchers' also added that prediction of crop N status was possible for all growth stages while P status could only be detected from v6 to v8 (when corn developed 6 to 8 leave including the first leaf) using blue (440-445 nm) and NIR (730-930 nm) bands.

CHAPTER III

MATERIALS AND METHODS

One experiment was conducted for two years (2008 and 2009) at Perkins, OK; 35^0.59'.55"N and 97^0.02'53"W, at an altitude of approximately 274 m (900 ft) above sea-level. The site has annual rainfall of 88.9 cm. The soil at this site is Konawa fine sandy loam with a pH of 6.1. The experimental design was a randomized complete block with three replications. The treatment structure contains twelve factorial treatment combinations of four N rates (0, 56, 112 and 168 kg N ha^{-1}) as Urea (46% N) and three P rates (0, 34 and 67 kg P ha^{-1}) as triple super phosphate (20% P). The plot size was 3.05 by 9.2 m with 3.05 m alleys. Winter wheat varieties; *Fannin* was planted on October 20, 2007 and *Duster* was planted on October 21, 2008 in a row spacing of 15 cm with seeding rates of 68.3 and 89.6 kg ha^{-1}, respectively.

Spectral measurements with spectrometer

Spectral measurements in each plot were taken at Feekes growth stages; 4 (beginning of the erection of the pseudo-stem, leaf sheaths beginning to lengthen), 5 (pseudo-stem formed by sheaths of leaves strongly erected), and 10 (sheath of last leaf completely grown out, ear swollen but not yet visible) in 2008. In 2009, spectral measurements were collected at Feekes 4, Feekes 7 (node of stem formed, next-to-last leaf just visible) and Feekes 10. Spectrometer measurements were taken using an Ocean Optics spectrometer 4000 (Ocean Optics Inc, Dunedin, FL) that operates in the range of 250 to 1200 nm wavelengths of the visible and NIR region with an analog to digital converter resolution of 16 bit and optical resolution of 1.5 nm full width half maximum (FWHM). A 2 m long glass fiber (Qp-1000-2-UV/VIS Ocean Optics Inc) with a diameter of 200 nm was connected to the spectrometer and the spectrometer was connected to a laptop computer that had Ocean Optics OIBase software which records the light intensity for each wavelength. This instrument has the capacity of taking 3648 pixels (the smallest unit in the picture that indicates the brightness of the color) at a time with a pixel size of 8 μm by 200 μm.

White plate ($BaSO_4$) reflection correction was used at all sites before readings were collected. Next to this, light reflected from white board was measured. Depending on time of the day, strength of solar radiation, and number of treatments, white board measurements were collected at different intervals to reduce intensity variability that hit the crop canopy.

Spectral data analysis

From the collected intensity at each wavelength, only intensities at wavelengths from 400 to 1000 nm were divided by the white board intensity to determine reflectance. Finally the reflectance was partitioned into 60 wavelengths at 10 nm bandwidths.

Spectral reflectances at each of the 60 spectral bands were correlated with forage P content. Similarly, analysis of variance was employed to determine the effect of N and P rates on each reflectance data. Based on their significance ($P \leq 0.05$) with forage N and P content and rate effect of N and P, reflectance at some wavelengths were selected to develop indices that can detect P deficiency in winter wheat. Indices were calculated as the ratio of the difference of NIR and visible, and the sum of NIR and the visible [(NIR - visible)/ (NIR + visible)]. This equation normalizes the value to -1 to +1 and results in indices similar to NDVI. However, for green plants, the index value must be a positive because much of the light in the visible portion of the electromagnetic spectrum is absorbed while much of the NIR spectra are reflected from the plant canopy. Three indices (Spectral Phosphorus Index (SPI) calculated as SPI1 [(915 - 455) / (915 + 455)], and SPI2 [(865 - 505) / (865 + 505)], and SPI3 [(915-495)/ (915+495)]) were developed from the combination of visible and NIR spectra.

Finally, a stepwise regression was used to identify suitable reflectance measurements and indices that were better related to N and P status of winter wheat crop. Decision for entering and removing variables was made using $p<0.15$. Final spectral reflectance measurements and indices were selected based on F-test and the partial regression sum of squares for each variable (Kutner et al., 2004).

Digital pictures

The contribution of digital pictures and image analysis software were not less in studying wheat crop (*Triticum aestivum L)* ground coverage (Purcell, 2000). Due to the indicated importance, digital pictures were taken at each of the above growth stages, using a Digital Olympus camera with a 6.0 mega pixel resolution, model number FE140, DC-3v, J69263481 which has 6.3 to 18.9 mm zooming capacity (Olympus imaging corporation, Indonesia). The digital pictures were converted to statistical values with the help of digital picture conversion software GNU image manipulation (GIMP – The GIMP team, 2001 - 2009) and the statistical values were manually collected from the picture gray scale. The gray scale enables collection of data from each pixel based on the mean color value of the red, green, and blue (RGB) colors and rating from 0 to 255 on the frequency histogram. From the above picture colors: three indices were developed (R/G, B/R, and B/G). However, these indices were negatively correlated with NDVI.

Greenseeker™ Sensor

The Greenseeker™ optical sensor that measures NDVI is active or self-illuminated and senses best at the height of 1 m from the sample crop. The device measures the fraction of reflected red and NIR radiation from the sample crop to the sensor. The NDVI measures the proportion of red and NIR reflectance from the crop canopy which is calculated as the ratio of the difference between NIR and red to the sum of red and NIR wavelengths (Raun et al. ,2001) and yields a value between 0 and 1 (Rouse et al., 1973). The higher the value, the more the crop is green and healthy. The Greenseeker™ Hand Held Optical sensor internally processes data and provides NDVI values. Details of the operation of this device have been discussed in the publication of Freeman et al. (2007) and Martin et al. (2007).

New-experimental 4 band sensor

The other instrument used for this work was the new-experimental-4-band (NEFB) handheld sensor which was equipped with active illumination similar to Greenseeker™ sensor. This sensor collects measurements at four wavelengths. One of the wavelengths was in the visible portion (660 nm) and the remaining three were in the NIR portion (780, 870, and 970 nm) of the electromagnetic spectrum. From these four wavelengths, three indices; were developed. The indices were calculated using the following equation: NEFB1 [(780-660) / (780+660)], NEFB2 [(870-660)/ (870+660)], and NEFB3 [(970-660)/ (970+660)].

Forage, grain, and soil sample collection and analysis

Following spectral measurements, forage samples were clipped at ground level from 1 m^2 areas, measured, and oven dried at 79 ^{0}C for 7 days. The dried forage samples were weighed and ground for forage N and P content analysis. At maturity, winter wheat was manually harvested from 1 m^2 areas using hand sickles. Grain was separated from the straw using a portable thrasher. The collected grain yield was weighed and then allowed to dry at 79 ^{0}C for 7 days. Dried samples were weighed and moisture content was determined from dried and wet grain. Yield was then adjusted to 12% standard grain moisture content. The dried samples were ground and processed to determine grain N and P content.

Postharvest composite soil samples were collected from 15 cores from each plot. The collected soil samples were allowed to dry in the air for 10 days. When it was dry enough, it was ground to pass through 2 mm sieve to separate the soil from voids.

The ground forage, grain, and soil were analyzed to determine grain, forage, and soil N and P contents, and soil pH. Forage P content was extracted using nitric acid digestion method. Total N in forage and soil was quantified by dry combustion method using a LECO carbon/nitrogen analyzer. Soil P was extracted using the Mehlich III extractant.

Analysis of Variance was used to test the effect of N, P and N by P rates interaction on measured variables including spectral measurements, and its single degree of freedom contrasts were calculated using the General linear model procedure (GLM) in SAS (SAS Institute, 2001). Correlation analysis was used to explore the relationship between sensor values and the forage and grain yield, and forage and grain N and P.

CHAPTER IV

RESULTS AND DISCUSSION

Spectrometer and winter wheat N and P status

Correlation analysis was employed for separate wavelength reflectance vs. forage and grain N and P content, and forage and grain yield. Except forage P, all remaining variables were negatively correlated with the reflectance in the visible portion of the spectrum starting from 405 to 715 nm and positively correlated with the NIR portion from 735 to 945 nm at 10 nm intervals. But the reflectance at 725 nm was not correlated with any of the variables in this study; rather it showed a non-significant transition point from negative to positive significant correlation for all variables except forage P content. Since, forage and grain N and P content, and grain yield correlated with the reading collected at different growth stages, the result of the analysis was based on the health status of the crop. Negative correlation of those variables with spectral indices in the visible region of the spectrum means, less reflectance of the red as tissue concentration of N increased. On the other hand, much of the NIR was reflected, so that it had a positive correlation with N tissue concentration. The opposite was true for forage N concentration.

From the above range of wavelengths; reflectance from 605 nm to 695 nm were consistently correlated with forage P content at Feekes 10 in 2008, and Feekes 7 and 10 in 2009. In the visible region of the spectrum, forage P content was positively correlated with separate wavelengths while negatively correlated in the NIR region of the spectrum. However, there were no significant correlations between separate wavelengths and forage P independent of the influence of forage N content. Reflectance measurements from 455 to 715 nm, and 815 to 855 nm wavelengths were correlated with forage N at Feekes 10 in 2008, and Feekes 7 and 10 in 2009. Similarly reflectance from 575 nm to 705 nm wavelengths were significant for forage yield at Feekes 5 and 10 in 2008, and Feekes 7 and 10 in 2009. The remaining reflectance measurements at different wavelengths were not consistently correlated with the variables at all growth stages but significant only at specific growth stages. No significant reflectance measurements at any wavelength detected winter wheat N and P nutrient status at Feekes 4 in 2008.

However, at the remaining growth stages and years, forage P content was significantly correlated with reflectance measurements that were significant for forage N content. The range of the wavelengths where reflectance was significantly correlated with forage N was wide and that encumbered the wavelengths that can detect forage P. For example, the wavelengths from 455 nm to 715 nm, and 815 nm to 855 nm at an average of 10 nm band width was significant ($P < 0.05$) for forage N content at Feekes 10 in 2008, and Feekes 7 and 10 in 2009, but reflectance at wavelengths from 605 nm to 695 nm (only the visible portion of the wavelength) was consistently significant for forage P content at the same growth stages in both years. When indices were developed for forage P content, at least one of the wavelengths was from the wavelength significant for forage

N content and detects both forage N and P content. In most cases it detected N better than P especially at late growth stages.

Also, reflectances at some wavelengths were correlated with grain yield, grain P, and grain N. Reflectance at wavelengths 745 to 925 nm was significantly correlated with grain yield at Feekes 5, and 10 in 2008 and Feekes 7 in 2009. From the reflectance of this region, the reading at 825 nm was more correlated with grain yield at Feekes 5 ($r = 0.47$, $P < 0.01$) and Feekes 10 ($r = 0.72$, $P < 0.001$) in 2008, and Feekes 7 ($r = 0.52$). At Feekes 7 in 2009, the visible portion of the electromagnetic spectrum (405 nm to 705 nm) was significant for grain yield with the highest significance level ($r = 0.72$, $P < 0.001$) at 415 nm. However, many wavelengths that were significant to predict grain yield and grain N content were not consistent at different growth stages of the same year or different years. Grain P was significantly correlated with reflectance from 735 to 985 nm wavelengths at Feekes 10 growth stage in 2008 with maximum significance ($r = 0.42$, $P < 0.05$) at 775 nm (Table 3).

From the stepwise regression analysis, there was no wavelength or index that was significant for measured variables at $P < 0.15$ significance level at Feekes 4 growth stages in 2008. At the remaining growth stages, inclusion of different reflectance measurements at different wavelengths in a model resulted in higher coefficient of determination value ($R^2 \leq 0.93$) than reflectance measurements at a single wavelength or ratio. Although high R^2 values were found for reflectance measurements at different wavelengths, they were not consistent across growth stages and years (Table 8 a, b, c, and d).

Combined wavelengths were developed as an equation from the reflectance of single wavelengths averaged from 10 nm band width and statistically tested for winter wheat forage P status. All non significant and non consistent equations were dropped. Three indices: Spectral Phosphorus Index (SPI) calculated as SPI1 [(915 - 455) / (915 + 455)], and SPI2 [(865 - 505) / (865 + 505)], SPI3 [(915-495)/(915+495)] were found to be consistent in detecting winter wheat forage P and N status over the two-year study. Spectral phosphorus index (SPI1) was significant for forage P content at Feekes 5 ($r = 0.5$, $P < 0.01$) and Feekes 10 ($r = - 0.35$, $P < 0.05$,) in 2008, and Feekes 10 ($r = -0.47$, $P < 0.01$) in 2009. Likewise, SPI2 was significantly correlated with forage P content at Feekes 5 ($r = 0.44$, $P < 0.01$) and Feekes 10 ($r = -0.36$, $P < 0.05$) in 2008, and Feekes 7 ($r = -0.32$, $P < 0.01$) and Feekes 10 ($r = - 0.57$, $P < 0.001$) in 2009. However, at Feekes 4, forage P content was not significant in both years.

Similarly, forage N was significantly correlated with SPI1 at Feekes 5 ($r = 0.46$, $P < 0.01$) and Feekes 10 ($r = 0.55$, $P < 0.001$) in 2008, and Feekes 7 ($r = 0.53$, $P < 0.01$) and Feekes 10 (r 0.40, $P < 0.05$) in 2009. Also, SPI2 was significantly correlated with forage N content at Feekes 5 ($r = 0.45$, $P < 0.01$) and Feekes 10 ($r = 0.63$, $P < 0.001$) in 2008, and Feekes 7 ($r = 0.5$, $P < 0.001$), and Feekes 10 ($r = 0.54$, $P < 0.001$) in 2009. So, the result from correlation analysis showed that an increase in forage P content decreased the value of SPI1 and SPI2 at Feekes 7 and 10 while an increase in forage N content increased the value of these indices at all growth stages. As a result, a negative significant relationship was observed between indices (SPI1 and SPI2) and forage P content at

Feekes 10 in 2008, and Feekes 7 and 10 in 2009 while it was positive and significant for forage N content (Table 4).

The overall trend showed a significant increase in forage P content increase the values of the spectral indices (SPI1 and SPI2) at Feekes 5 in 2008, but decreased while forage P content increased at Feekes 7, 2009 and Feekes 10 for both years. This trend was as a result of crop growth stage that increased crop biomass (Figure 1a and 2a) and decreased forage P content (Figure 1b and 2b) due to N fertilization and biomass dilution effect. For example, the mean forage yield harvested at Feekes 10 in 2008 from 67 kg P ha^{-1} treatment was 3.1 Mg ha^{-1} and from 168 kg N ha^{-1} by 67 kg P ha^{-1} treatment was 11.1 Mg ha^{-1}. Mean forage P content found at 67 kg P ha^{-1} was 2.6 g kg^{-1} while it was 2.4 g kg^{-1} from 168 kg N ha^{-1} by 67 kg P ha^{-1} treatment. Treatments that yielded less biomass showed high forage P content compared to treatments that yielded high biomass.

Analysis of Variance showed that except N rate, there was no significant effect of P rate on any of the separate wavelengths for both years. From the whole range of wavelengths where spectral reflectance was significantly affected by N rate, only those with model R^2 values greater than or equal to 0.65 were selected (Table 9 a, b, c). Also, the reflectance at wavelengths from 655 to 695 nm were significantly affected by N rate at Feekes growth stages 10 in 2008 and Feekes 4,7,10 in 2009 with maximum model R^2 values and $P < 0.001$ at each growth stage. At Feekes 10 in 2008, only linear N rate affected reflectance from 465 to 695 nm. However, reflectance at 485 and 495 nm were not included in this result, because their corresponding model R^2 values were less than

0.65. Similarly in 2009, reflectance from wavelengths: 565 nm to 695 nm at Feekes 4; 445 nm to 705 nm, and 745 nm to 915 nm at Feekes 7; and from 655 to 695 nm at Feekes 10 were significantly affected by N rate. Except reflectance at wavelengths; 525, 555, and 675 nm at Feekes 7, and 675 nm at Feekes 10 in 2009, both linear and quadratic contrast effect of N rate were significant ($P < 0.001$).

Analysis of variance showed that P rate affected forage P content but did not affect SPI1 and SPI2 at Feekes 4 in 2008. However, SPI2 was significantly affected by P rate at Feekes 5 (Table 5 b) in 2008 and Feekes 4 (Table 5 d) in 2009. Phosphorus rate did not affect both indices at Feekes 7 (Table 5 e) in 2009 and Feekes 10 (Table 5 c and f) in both years. Osborne et al. (2002) reported that spectral measurements passed v8 growth stage in corn were not important in predicting forage P content while before v8 were useful using the wavelengths (440 nm and 445 nm) and (730 nm and 930 nm) while N was predicted throughout corn growth stages. Therefore, spectral detection of winter wheat forage P and N content had similar characteristics with corn during late growth stages when SPI1 and SPI2 were used for winter wheat.

In this study there was no effect of P rate on the spectral indices independent of N and N by P rate interaction. Thus, this result supported the result found from the correlation analysis of reflectance at separate wavelengths and the value of SPI1 and SPI2 vs. forage P content except the R/G index. Furthermore, there was no significant N by P rate interaction effect on SPI1 and SPI2 in 2008 but the effect was significant at Feekes 4 and 7 in 2009. The spectral measurements taken at Feekes 7 in 2008 and Feekes

5 in 2009 were not included in the analysis because of the influence of uncontrolled weather condition (cloud cover, and rain fall) during data collection.

Spectral value, forage & grain yield, and grain N & P

The effect of P rate on biomass gradually decreased towards at later growth stages unlike N rate. Similar results reported from a corn study showed that applied P did not significantly affect biomass at later growth stages (Osborne et al., 2002). In our study, forage yield was significantly correlated with the value of SPI 1 and 2 at all growth stages excluding Feekes 4 in 2009 when there was no sample from P treatments (Table 4). Spectral Phosphorus Index 1 (SPI1) was significantly correlated with forage yield at Feekes 4 ($r = 0.45$, $P < 0.01$), Feekes 5 ($r = 0.68$, $P < 0.001$), Feekes 10 ($r = 0.9$, $P < 0.001$) in 2008, and Feekes 7 ($r = 0.90$, $P < 0.001$) and Feekes 10 ($r = 0.37$, $P < 0.001$) in 2009. For SPI 2, correlation analysis also showed similar results at Feekes 4 ($r = 0.45$, $P < 0.01$), Feekes 5 ($r = 0.63$, $P < 0.001$), Feekes 10 ($r = 0.93$, $P < 0.001$) in 2008, and Feekes 7 ($r = 0.89$, $P < 0.001$) and Feekes 10 ($r = 0.52$, $P < 0.001$) in 2009 (Table 4).

Spectral reflectance collected at Feekes growth stages 5 and 10 in 2008, and Feekes 7 and 10 in 2009 were correlated with grain yield and grain N (Table 3). This finding supported the results of ANOVA that N rate affected grain yield and grain N content (Table 6). On the other hand, since P rate affected grain P only at Feekes 10 in 2009 (Table 6), there was no significant wavelength that was consistently correlated with grain P. Moreover, forage P content was significantly correlated with grain yield at

Feekes 4 ($r = 0.35$, $P < 0.05$) in 2008 and Feekes 10 ($r = 0.51$, $P < 0.01$) in 2009 but was not significant with grain P at all growth stages of the two years (Table 7).

Picture Index

Three indices were developed from the collected picture statistical value and statistically evaluated for their correlation with different variables included in the study. Two indices (R/G and B/R) were correlated with several variables (Table 4). A consistent significant correlation was observed between (R/G) index, and forage yield and forage P content in 2008. This index was significant for forage yield, at Feekes 4 ($r = -0.90$, $P < 0.001$), Feekes 5 ($r = -0.66$, $P < 0.001$), and Feekes 10 ($r = -0.81$, $P < 0.001$) in 2008, and at Feekes 7 ($r = -0.48$, $P < 0.01$) in 2009. Forage P content was significantly correlated with this index only at Feekes 4 ($r = -0.66$, $P < 0.001$) and Feekes 5 ($r = -0.49$, $P < 0.01$) in 2008. However, the index was not significant for forage N content overall growth stages.

Picture index B/R was significantiy correlated with forage N content only at Feekes 5 ($r = 0.74$, $P < 0.001$) in 2008 and Feekes 7 ($r = 0.56$, $P < 0.001$) in 2009. This index was significant for forage yield at all growth stages: Feekes 4 ($r = 0.37$, $P < 0.05$), Feekes 5 ($r = 0.6$, $P < 0.001$) and Feekes 10 ($r = -0.67$), $P < 0.001$) in 2008, and Feekes 7 ($r = 0.63$, $P < 0.001$) and Feekes 10 ($r = -0.36$, $P < 0.05$) in 2009. The correlation between forage P content and the index B/R was significant at Feekes 5 ($r = 0.37$, $P < 0.05$) and Feekes10 ($r = 0.34$, $P < 0.05$) in 2008, and Feekes 10 ($r = 0.41$, $P < 0.05$) in 2009.

The value from picture index R/G was inversly related at earlier growth stages (Feekes 4,5, and 7) with other indices for forage yield because it was developed based on the proportion of mean red to mean green reading of the picture gray scale. So, the lesser the red and the more the green, the lesser the ratio which was negativelly correlated with high and green biomass. Over all, from the picture indices (R/G, B/R and B/G), R/G showed more promissing result in indicating winter wheat forage P content independent of forage N content.

The Greenseeker™ sensor NDVI

The NDVI from Greenseeker™ sensor was significantly correlated with forage yield, and forage N and P contents at all growth stages of winter wheat except for forage N content at Feekes 4 in 2008. Also, forage yield was significant with NDVI at Feekes 4 ($r = 0.88$, $P < 0.001$), Feekes 5 ($r = 0.84$, $P < 0.001$), and Feekes 10 ($r = 0.92$, $P < 0.001$) in 2008 and Feekes 7 ($r = 0.77$, $P < 0.001$) in 2009. Forage P content was significant at Feekes 4 ($r = 0.69$, $P < 0.001$), Feekes 5 ($r = 0.43$, $P < 0.01$), and Feekes 10 ($r = -0.37$, $P < 0.05$) in 2008. At Feekes 10, the correlation between NDVI and forage P content was negative and significant while it was positive at the remaining Feekes growth stages like SPI1 and SPI2. However the correlation between forage P content and NDVI was not significant at Feekes 7 and 10 in 2009. Forage N was significantly correlated with NDVI at Feekes 5 ($r = 0.77$, $P < 0.001$), and Feekes 10 ($r = 0.52$, $P < 0.01$) in 2008, and Feekes 7 ($r = 0.39$, $P < 0.05$) in 2009 (Table 4).

Analysis of variance showed that NDVI was significantly affected by N rate at Feekes 4 (Table 5 a), Feekes 5 (Table 5 b), and Feekes 10 (Table 5 c) in 2008, and Feekes 7 (Table 5 e) in 2009. The index was also affected by P rate at Feekes 4 in 2008 and Feekes 7 in 2009.

New-experimental 4 band sensor index

From the reflectance of the NEFB sensor; neither the reflectance value from separate wavelengths nor from developed indices of the NEFB sensor were significantly correlated with winter wheat forage yield and forage N and P content at Feekes 4 in 2008. The index NEFB1 was significantly correlated with forage yield at Feekes 5 ($r = 0.52$, $P < 0.01$) and Feekes 10 ($r = 0.87$, $P < 0.001$) in 2008, and Feekes 7 ($r = 0.77$, $P < 0.001$) in 2009. The index NEFB2 had almost the same correlation coefficient value and probability level with NEFB1 index for forage yield at Feekes 5 and 10 in 2008, and Feekes 7 and 10 in 2009. The index NEFB3 had a significant ($P \leq 0.05$) relationship with forage yield at the above mentioned growth stages except at Feekes 7 in 2009 (Table 4).

At Feekes 10 in 2008, forage P content was negative and significantly correlated with all indices of NEFB sensor. Similarly, forage P content was significant and negatively correlated with NEFB1 and NEFB2 at Feekes 7 in 2009. Forage N content was also significantly correlated with all the three indices at Feekes 5 ($r = 0.60$, $P < 0.001$) and Feekes 10 ($r = 0.53$, $P < 0.001$) in 2008 and at Feekes 7 ($r = 0.53$, $r= 54$, and $r= 55$, $P < 0.01$) for indices NEFB1, NEFB2 and NEFB3 respectively in 2009 (Table 4). These indices were significantly affected by N rate at Feekes growth stages 4, 5, and 10 in 2008, and Feekes 7 in 2009 (Table 5 a, b c, and e).

Forage P content at different growth stages

Forage P content was the lowest at booting (Feekes 10) growth stage (Figure 1b and 2 b) largely due to a rapid increase in crop biomass compared to Feekes 4 and 5 growth stages. For example, the percent forage yield at Feekes 4 was 9.4 of the biomass at Feekes 10 from the check plot while forage P content at Feekes 10 was 53 percent of the content at Feekes 4 in 2008. This shows that forage yield increased by 91.6 percent while forage P content decreased by 47 percent from Feekes 4 to 10. At Feekes 5, forage yield of this treatment was increased to 21 percent of the biomass at Feekes 10 but forage P content was 107 percent of the content at Feekes 4. The other example was from N and P rate applied treatment. At Feekes 4 the treatment with 168 kg N ha^{-1} and 67 kg P ha^{-1}, had a forage yield that was 8 percent of the forage yield at Feekes 10 but forage P content at Feekes 10 was 45 percent of the content at Feekes 4 in 2008. This shows that increase in growth stage increased forage yield and decreased forage P content.

Forage yield gradually increased as wheat continued to grow when sufficient N was applied. This increase in forage yield was as a result of increased size and number of roots that were competing for P more than what the crop supplied from the soil. So, P in the forage was redistributed throughout the growing plant and reduced the overall forage P content. Although forage P content was decreased over growth stages, it increased at each growth stage as P rate applied was increased. At Feekes 4, 5, and 10, forage P content increased as applied P rate increased (Figure 1b and 2b), and more increased with N and P rate applied than P rate alone at Feekes 4 and 5. For example, mean forage P content at Feekes 4 in 2008 was increased by 14 percent over the check to 67 kg P ha^{-1}

and increased by 44 percent over the check to 168 N and 67 P kg ha^{-1}. Similar results have been documented on the variation of corn and spring wheat shoot N and P content as a result of N fertilization (Ziadi et al., 2007; 2008).

Moreover, from 2008 and 2009 post harvest soil and forage P analysis, applied P significantly affected forage P content (Table 5 a, b, c, e and f) and residual soil P level (Table 6). Residual soil P level was increased from 2008 to 2009 even for the check plot (Figure 3).

CHAPTER V

CONCLUSIONS

Reflectance at several bands were significantly correlated with forage N and P content at Feekes growth stages 5, 7 and 10. However, all wavelengths that detected forage P content were within the range of the wavelengths that were significantly correlated with forage N content. Reflectance at these wavelengths was consistently correlated with forage N and P content, and forage yield at several growth stages during the study period. Analysis of variance showed that there was no significant effect of P rate on reflectance of separate wavelengths at all growth stages over the two years.

Consistent correlation had been observed from SPI1 and SPI2 in identifying winter wheat forage P content. These indices plus NDVI, and NEFB1, NEFB2 & NEFB3 indicies were negatively correlated with forage P and positively with forage N contents at later growth stages arround Feekes 7 to Feekes 10. This was likely due to the dilution effect of the biomass as a result of N fertilization and increasing in crop growth stage. Contrary to this, the relationship between forage P content and the indicated indices was positive at an earlier growth stage (Feekes 5) of winter wheat. According to this finding the only picture index that had a promising potential to identify forage P content independent of forage N content was the R/G index. Forage yield had strong and consistent correlation with SPI1 at all growth stages over two years.

In this work, three general properties of forage P content were observed.

1) As the rate of applied P increased, forage P content increased. 2) As rate of N and P applied increased, forage P content was increased more than P rate alone at Feekes 4 and 5. 3) As biomass increased, forage P content decreased over growth stages because of the biomass dilution effect. Analysis of variance showed that rate of applied P affected soil residual P analysed from post harvest soil samples. This may limit forage P content increase based on rate applied at later growth stages (Feekes 7 and above) and may have an effect on spectral reflectance values.

In general, this work confirmed the possibility of developing consistent indices that can detect winter wheat forage P status. However, more test data is needed to evaluate the usefulness of the spectral and picture indices we have developed. Our results showed that the correlation between forage P content and SPIs were changed from positive to negative as the season progressed from Feekes 5 to 8. Therefore, additional research is needed to determine the specific growth stage at which P content starts to relate negatively with SPIs. Also, it is important to recognize extreme P deficient winter wheat forage response to SPI1 and SPI2 particularly at later growth stages (After Feekes 7 wheat growth stage).

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1. Table 1. Initial surface (0 – 15cm) soil test characteristics of the experimental site, Teller sandy loam soil, Perkins, OK, 1998.

Characteristics	Method	Unit	Soil test level	Critical level
pH	1:1 soil:H_20	-	5.9	5.7
Organic Carbon†	Dry Combustion	g kg^{-1}	5.336	
Total Nitrogen†	Dry Combustion	g kg^{-1}	0.504	
NH_4-N‡	2 M KCl extract	mg kg^{-1}	3.0	
NO_3-N‡	2 M KCl extract	mg kg^{-1}	2.8	40
Phosphorus§	Mehlich-3	mg kg^{-1}	8.9	32.5
Potassium§	Mehlich-3	mg kg^{-1}	133.0	125

†Schepers et al. (1989)
‡Lachat instruments (1989)
§Mehlich (1984)

Table 2. Experiment site, years and days of planting and data collection, Perkins, OK, 2008-2009

Year	**Planting date**	**Data collection**	**Growth stages**
2007-2008	10-21-2007	02 – 28 - 2008	Feekes 4
		03 - 21-2008	Feekes 5
		05 - 01- 2008	Feekes 10
		06 - 12 - 2008	Harvest
		06 - 20 - 2008	Postharvest soil sample
2008-2009	10-20-2008	02 - 26 - 2009	Feekes 4
		03 - 30 - 2009	Feekes7
		04 - 14 - 2009	Feekes 10
		06 - 10 - 2009	Harvest
		06 - 13 - 2009	Postharvest soil sample

Table 3. Selected range of 10 nm bandwidths that were significantly correlated with forage N and P, content, forage and grain yield, grain N and P content at Feekes 5, 7 and 10, Perkins, OK, 2008 -2009

Year	Variables	Growthstages		
		Feekes 4	Feekes 5	Feekes 10
2008	Forage P content	-	-	445-695
	TissueN	-	-	(455-715),(815-925)
	Forage yield	-	575-705	(445-715),(735-975)
	Grain yield	405-705	745-925	(445-705),(745-935)
	Grain N	-	745-905	(415-705
	Grain P	-	-	(735-985)
2009		Feekes 4	Feekes 7	Feekes 10
	Forage P content	-	495-695	(435-515),(575-695),(745-785)
	TissueN	-	(405-715) (735-925)	(435-715)(715-855)
	Forage yield	-	(405-715)(735-935)	435-715
	Grain yield	-	405-715	435-715
	Grain N	(575-705) (735-935)	405-715	435-715
	Grain P	-	-	-

Numbers in the table indicate range of wavelengths significant at P< 0.05 probability level depending on the type of the variable. E. g. 575 - 685 would include 575 nm, 585 nm, 595 nm, up to 695 at an average of 10 nm band width.

Table 4. Correlation of indices with the picture index, New-experimental 4 band sensor, and NDVI with forage yield, forage N and P content at Feekes 4, 5, 7 and 10 growths stages, Perkins, OK, 2008 - 2009.

Year	**Indices**	**Feekes 4**			**Feekes 5**			**Feekes 10**		
		Forage yield	Forage P	Forage N	Forage yield	Forage P	Forage N	Forage yield	Forage P	Forage N
2008	SPI 1	.45 **	NS	NS	.68***	.50**	.45**	.90***	-.35*	.41*
	SPI 2	.45**	NS	NS	.63***	.44**	.45**	.93***	-.36*	.63**
	SPI 3	.43*	NS	NS	.62***	.44**	.42**	NS	NS	NS
	R/G	-.90***	-.66***	NS	-.66***	-.49**	NS	-.81***	NS	NS
	B/R	.37*	NS	NS	.60***	.37*	.74***	-.67***	.34*	NS
	B/G	NS	NS	NS	-.43**	NS	NS	-.77***	.34*	NS
	NDVI	.88***	.69***	NS	.84***	.43**	.77***	.92***	-.37*	.52**
	NEFB 1	NS	NS	NS	.52**	NS	.60***	.87***	-.42**	.53***
	NEFB 2	NS	NS	NS	.51**	NS	.60***	.88***	-.42**	.54***
	NEFB 3	NS	NS	NS	.51**	NS	.60***	.86***	-.42*	.55***
2009		Feekes 4			Feekes 7			Feekes 10		
		Forage yield	Forage P	Forage N	Forage yield	Forage P	Forage N	Forage yield	Forage P	Forage N
	SPI 1	-	-	-	.90***	NS	.53**	.37***	-.47**	.40*
	SPI 2	-	-	-	.89***	-.32**	.50***	.75***	-.57***	.54***
	SPI 3	-	-	-	.78***	-.33*	.51**	.80***	.56***	.66***
	R/G	-	-	-	-.48**	NS	NS	NS	NS	NS
	B/R	-	-	-	.63***	NS	.56***	-.36*	.41*	NS
	B/G	-	-	-	NS	NS	NS	-.38*	.39*	NS
	NDVI	-	-	-	.77***	NS	.39*	NS	NS	NS
	NEFB 1	-	-	-	.75***	-.33*	.53**	.37*	NS	NS
	NEFB 2	-	-	-	.76***	-.32*	.54**	.37*	NS	NS
	NEFB 3	-	-	-	NS	NS	.55***	.35*	NS	NS

- NS,*, **, ***, Not significant, significant <0.05, <0.01, <0.001, respectively with their correlation coefficient (r) values; forage N or P = forage N or P content
- Phosphorus Index (SPI) SPI1 [(915 - 455) / (915 + 455)], SPI2 [(865 - 505) / (865 + 505)], and SPI3 [(915-495)/(915+495)]
- New Experimental Four Band (NEFB); NEFB1 [(780-660) / (780+660)], NEFB2 [(870-660)/ (870+660)], and NEFB3 [(970-660)/ (970+660)].

Table 5a. Analysis of variance, single degree of freedom contrasts, treatment mean squares, and treatment means for forage yield, forage N and P content, and spectral, New-experimental 4 band sensor and, digital picture indices at Feekes 4 growth stages, Perkins, OK, 2008

	Source of variation	DF	Forage yield Mg ha^{-1}	Forage P g kg^{-1}	Forage N g kg^{-1}	NDVI	Spectral indices			New experimental 4-band indices			Digital Picture indices	
							SPI 1	SPI 2	SPI 3	NEFB 1	NEFB 2	NEFB 3	R/G	B/R
Feekes 4_2008	N rate	3	0.24***	.017***	NS	.038***	NS	NS	NS	0.016*	0.011*	0.09*	.005**	NS
	P rate	2	**0.24*** **	.109***	NS	.029***	NS	NS	NS	NS	NS	NS	.01***	NS
	N rate*P rate	6	NS	.012***	NS	NS	NS	NS	NS	NS	NS	NS	.004**	NS
	R-square	-	**0.78**	0.89	0.2	0.85	0.47	0.40	0.39	0.45	0.45	0.45	0.76	0.4
	Contrast													
	Linear N rate	1	.686 ***	.045***	NS	.111 ***	NS	NS	NS	.0474**	.031**	.026**	.016***	NS
	Quadratic N rate	1	NS	NS	NS	.057 ***	NS	NS	NS	NS	NS	NS	NS	NS
	Linear Prate cont	1	.406 ***	.213***	NS	NS	NS	NS	NS	NS	NS	NS	.02 ***	NS
	Quadratic Prate	1	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
	Linear N*P	1	.223 **	.066***	NS	.011*	NS	NS	NS	NS	NS	NS	.014***	NS
	Treatment Means													
	N rate, kg ha-1	0	0.322	2.90	38.69	0.294	0.281	0.451	0.359	0.39	0.457	0.477	0.960	0.717
		56	0.470	3.52	36.99	0.328	0.317	0.510	0.402	0.384	0.456	0.476	0.937	0.761
		112	0.626	3.53	37.04	0.407	0.324	0.519	0.414	0.421	0.483	0.5	0.922	0.793
		168	0.682	3.93	36.10	0.433	0.329	0.529	0.420	0.431	0.493	0.51	0.902	0.792
		SED	0.18	0.311		0.094				0.117	0.110	0.110	0.083	
	P rate, kg ha-1	0	0.364	2.44	37.54	0.346	0.30	0.481	0.383	0.381	0.449	0.47	0.961	0.761
		34	0.587	3.64	36.25	0.367	0.324	0.516	0.411	0.393	0.459	0.48	0.93	0.763
		67	0.624	4.34	37.81	0.413	0.314	0.51	0.403	0.445	0.508	0.524	0.902	0.774
		SED	0.155	0.27		0.082				0.102	0.096	0.092	0.072	

- NS,*, **, ***, not significant, significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively with their mean square values.
- SED – standard error of the difference for two equally replicated means; forage N or P = forage N or P content.
- Phosphorus Index (SPI) SPI1 [(915 - 455) / (915 + 455)], SPI2 [(865 - 505) / (865 + 505)], and SPI3 [(915-495)/(915+495)]
- New Experimental Four Band (NEFB); NEFB1 [(780-660) / (780+660)], NEFB2 [(870-660)/ (870+660)], and NEFB3 [(970-660)/ (970+660)]

Table 5 b. Analysis of variance, single degree of freedom contrasts, treatment means squares, and treatment means for forage yield, forage N and P content, and spectral, New-experimental 4 band sensor and digital picture indices at Feekes 5 growth stages, Perkins, OK, 2008

e	**Source of variation**	DF	Forage Mg ha-1	Forage P g kg-1	Forage N g kg-1	NDVI	Spectral indices			New experimental 4-band indices			Digital Picture indices	
							SPI 1	SPI 2	SPI 3	NEFB 1	NEFB 2	NEFB 3	R/G	B/R
Feekes 5__ 2008	N rate	3	2.77***	.004*	2.95***	.160***	.005**	.016**	.008**	.03**	.021**	.020**	.002**	NS
	P rate	2	1.25*	.116***	.121*	NS	.004*	.010*	.006*	NS	NS	NS	0.002*	NS
	N *P rate	6	NS	0.006***	NS	NS	NS	NS	NS	NS	NS	NS	.0013*	NS
	R-square	-	.64	.93	.93	.78	.71	.71	.70	.58	.58	.58	.71	79
	Contrast													
	Linear N rate	1	8.25***	.007*	8.67***	.467***	.014***	.045***	.021***	.09***	.05***	NS	.006***	.143***
	Quadratic N rate	1	NS	.005*	.1681*	NS	NS	NS	NS	NS	NS	NS	NS	.022**
	Linear Prate	1	1.94*	.22***	NS	0.042*	.008**	.02**	.012**	NS	NS	NS	.003**	NS
	Quadratic P rate	1	NS	.014***	.1605*	NS	NS	NS	NS	NS	NS	NS	NS	NS
	Linear N*P	1	NS	.031***	NS	NS	NS	NS	NS	.023*	NS	.016*	.002*	.024**
							Treatment Means							
	N rate, kg ha-1	0	0.777	3.290	22.744	0.423	0.202	0.308	0.234	0.461	0.522	0.532	0.969	0.680
		56	1.325	3.159	25.367	0.528	0.223	0.341	0.257	0.510	0.561	0.564	0.946	0.695
		112	1.676	3.314	30.356	0.663	0.223	0.353	0.262	0.558	0.607	0.607	0.941	0.739
		168	2.087	3.64	35.711	0.718	0.261	0.410	0.304	0.62	0.661	0.66	0.931	0.853
		SED	0.356	0.262	0.624	0.137	0.079	0.104	0.091	0139	0.132	0.126	0.068	0.106
	P rate, kg ha-1	0	1.094	2.260	29.600	0.537	0.21	0.325	0.242	0.553	0.602	0.603	0.961	0.729
		34	1.643	3.63	27.6	0.593	0.227	0.352	0.264	0.517	0.583	0.572	0.941	0.737
		67	1.663	4.163	28.433	0.62	0.246	0.382	0.287	0.543	0.593	0.597	0.938	0.76
		SED	0.301	0.228	0.542	0.119	0.068	0.09	0.079	0.120	0.114	0.11	0.059	0.092

- NS,*, **, ***, not significant, significant at P < 0.05, P < 0.01, P < 0.001, respectively with their mean square values.
- SED – standard error of the difference for two equally replicated means, forage N or P = forage N or P content.
- Phosphorus Index (SPI) SPI1 [(915 - 455) / (915 + 455)], SPI2 [(865 - 505) / (865 + 505)], and SPI3 [(915-495)/(915+495)]
- New Experimental Four Band (NEFB); NEFB1 [(780-660) / (780+660)], NEFB2 [(870-660)/ (870+660)], and NEFB3 [(970-660)/ (970+660)].

Table 5c. Analysis of variance, single degree of freedom contrasts, treatment mean squares, and treatment means for forage yield, forage N and P content, and spectral, New-experimental 4 band sensor and, digital picture indices at Feekes 10 growth stages, Perkins, OK, 2008

Feekes 10 2008 Source of variation	DF	Forage Mg ha^{-1}	Forage P g kg^{-1}	Forage N g kg^{-1}	NDVI	Spectral indices			New experimental 4-band indices			Digital Picture indices	
						SPI 1	SPI 2	SPI 3	NEFB 1	NEFB 2	NEFB 3	R/G	B/R
N rate	3	92.6***	.005***	.339**	.20***	.01***	.023***	NS	.11***	.077****	.059***	.072**	.041**
P rate	2	NS	.014***	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
N *P rate	6	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
R-square	-	.83	.76	.56	.85	.74	.80	.63	.82	.82	.79	.62	.62
Contrast													
Linear N rate	1	25***	.006**	.856**	.6***	.029 ***	.068 ***	Ns	.318***	.176***	.175***	.175***	.058**
Quadratic N rate	1	NS	.008**	NS	NS	NS	NS	NS	NS	NS	NS	NS	.0413*
Linear P rate	1	NS	.027***	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Quadratic P rate	1	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Linear N*P rate	1	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
						Treatment Means							
N rate, kg ha-1	0	3..244	2.008	6.933	0.338	0.277	0.4933	0.051	0.317	0.378	0.392	0.937	0.531
	56	6.433	1.578	6.884	0.451	0.299	0.536	0.027	0.401	0.458	0.460	0.906	0.473
	112	9.67	1.489	8.422	0.599	0.341	0.589	0.061	0.498	0.544	0.532	0.759	0.369
	168	10.144	1.622	11.011	0.670	0.347	0.606	0.059	0.575	0.613	0.601	0.778	0.447
	SED	0.623	0.244	0.542	0.126	0.075	0.082	0.097	0.118	0.115	0.12	0.139	0.133
P rate, kg ha-1	0	6.692	1.281	8.80	0.507	0.314	0.557	0.061	0.456	0.505	0.504	0.867	0.481
	34	7.67	1.766	8.082	0.523	0.321	0.557	0.057	0.448	0.499	0.497	0.823	0.433
	67	7.76	1.976	8.057	0.513	0.313	0.554	0.031	0.439	0.491	0.488	0.846	0.452
	SED	0.54	0.213	0.469	0.11	0.065	0.071	0.084	0.102	0.099	0.095	0.120	0.115

- NS,*, **, ***, not significant, significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively with their mean square values.
- SED – standard error of the difference for two equally replicated means, forage N or P = forage N or P content.
- Phosphorus Index (SPI) SPI1 [(915 - 455) / (915 + 455)], SPI2 [(865 - 505) / (865 + 505)], and SPI3 [(915-495)/(915+495)]
- New Experimental Four Band (NEFB); NEFB1 [(780-660) / (780+660)], NEFB2 [(870-660)/ (870+660)], and NEFB3 [(970-660)/ (970+660)]

Table 5d. Analysis of variance, single degree of freedom contrasts, treatment mean squares, and treatment means for forage yield, forage N and P content, and spectral New-experimental 4 band sensor and, digital picture indices at Feekes 4 growth stages, Perkins, OK, 2009

Growth stage	Source of variation	DF	Forage Mg ha^{-1}	Forage P g kg^{-1}	Forage N g kg^{-1}	NDVI	Spectral indices			New experimental 4-band indices			Digital Picture indices	
							SPI 1	SPI 2	SPI 3	NEFB 1	NEFB 2	NEFB 3	R/G	B/R
Fekes 4_2009	N rate	3	-	-	-	NS	.12***	.021***	.016***	NS	NS	NS	NS	NS
	P rate	2	-	-	-	NS	.001*	.021*	.002**	NS	NS	NS	NS	NS
	N *P rate	6				NS	.002**	.002**	.002**	NS	NS	NS	NS	NS
	R-square	-	-	-	-	-	.97	.94	.95	-	-	-	-	-
	Contrast													
	Linear N rate	1	-	-	-	NS	.265***	.047***	.035***	NS	Ns	Ns	NS	NS
	Quadratic Nate	1	-	-	-	NS	.0.009***	.018***	.013***	NS	NS	NS	NS	NS
	Linear Prate	1	-	-	-	NS	0.002**	.004**	.003**	NS	NS	NS	NS	NS
	Quadratic Prate	1	-	-	-	NS	NS	.001*	NS	NS	NS	NS	NS	NS
	Linear N*P rate	1	-	-	-	NS	0.008***	.009***	.001**	NS	NS	NS	NS	NS
							Treatment Means							
	N rate, kg ha-1	0	-	-	-	NS	0.30	0.435	0.407	0.428	0.507	0.542	0.916	0.847
		56	-	-	-	NS	0.348	0.505	0.469	0.468	0.542	0.573	0.926	0.787
		112				NS	0.385	0.547	0.504	0.444	0.519	0.550	0.938	0.795
		168	-	-	-	NS	0.370	0.529	0.489	0.441	0.515	0.551	0.956	0.794
		SED				0.016	0.06	0.07	0.063	0.16	0.146	0.137	0.132	0.12
	P rate, kg ha-1	0	-	-	-	NS	0.339	0.486	0.451	0.428	0.507	0.542	0.944	0.772
		34	-	-	-	NS	0.356	0.513	0.476	0.496	0.564	0.593	0.930	0.724
		67	-	-	-	NS	0.359	0.512	0.474	0.412	0.492	0.527	0.927	0.754
		SED				0.012	0.053	0.06	0.055	0.138	0.126	0.118	0.114	0.10

- NS,*, **, ***, not significant, significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively with their mean square values.
- SED – standard error of the difference for two equally replicated means, forage N or P = forage N or P content.
- Phosphorus Index (SPI) SPI1 [(915 - 455) / (915 + 455)], SPI2 [(865 - 505) / (865 + 505)], and SPI3 [(915-495)/(915+495)]
- New Experimental Four Band (NEFB); NEFB1 [(780-660) / (780+660)], NEFB2 [(870-660)/ (870+660)], and NEFB3 [(970-660)/ (970+660)].

Table 5e. Analysis of variance, single degree of freedom contrasts, treatment mean squares and treatment means for forage yield, forage N and P content, and spectral, New-experimental 4 band sensor and, digital picture indices at Feekes 7 growth stages, Perkins, OK, 2009

Source of variation	DF	Forage Mg ha^{-1}	Forage P g kg^{-1}	Forage N g kg^{-1}	NDVI	Spectral indices			New experimental 4-band indices			D Picture indices	
						SPI 1	SPI 2	SPI 3	NEFB 1	NEFB 2	NEFB 3	R/G	B/R
Feekes7 2009													
N rate	3	36.6***	.032**	.94*	.117***	.048***	.061***	.017**	.29***	.195***	.153***	.065***	.034***
P rate	2	22***	.015*	NS	.025**	NS	NS	NS	NS	NS	NS	NS	NS
N *P rate	6	NS	.012*	NS	.007*	.004*	.005*	NS	NS	NS	NS	NS	NS
R-square	-	.89	.70	.49	.89	.85	.84	.84	.86	.85	.85	.77	.71
Contrast													
linear N rate	1	75.1***	.023*	2.57**	.228***	.108***	.131***	.054***	.67***	.454***	.356***	.146***	.085***
Quadratic N rate	1	34.4***	.072***	NS	.124***	.035***	.052***	.117***	.201**	.13***	.100***	.048**	Ns
linear Prate	1	41.8***	.024*	NS	.031**	NS	NS	NS	NS	NS	NS	NS	NS
Quadratic P rate	1	NS	NS	NS	.020*	NS	NS	NS	NS		NS	NS	NS
linear N*Prate	1	27.9***	.029*	NS	.023**	.014**	NS	.012*	NS	.027*	0.021*	NS	NS
Treatment Means													
N rate, kg ha-1	0	0.391	0.397	1.824	0.240	0.411	0.538	0.507	0.382	0.483	0.519	1.01	0.707
	56	2.29	0.27	1.981	0.423	0.517	0.664	0.624	0.639	0.692	0.696	0.875	0.766
	112	3.01	0.271	2.323	0.500	0.575	0.723	0.68	0.782	0.811	0.808	0.822	0.843
	168	2.865	0.322	2.629	0.449	0.554	0.70	0.656	0.74	0.773	0.777	0.836	0.827
	SED	0.402	0.117	0.322	0.15	0.091	0.098	0.092	0.139	0.126	0.12	0.130	0.114
P rate, kg ha-1	0	1.667	0.274	2.297	0.350	0.5052	0.648	0.608	0.594	0.655	0.668	0.900	0.772
	34	2.37	0.334	2.157	0.437	0.529	0.672	0.632	0.667	0.716	0.723	0.877	0.781
	67	2.38	0.337	2.114	0.423	0.509	0.649	0.611	0.646	0.700	0.709	0.878	0.802
	SED	0.349	0.1	0.28	0.092	0.071	0.085	0.081	0.12	0.11	0.104	0.113	0.098

- NS,*, **, ***, not significant, significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively with their mean square values.
- SED – standard error of the difference for two equally replicated means, forage N or P = forage N or P content.
- Phosphorus Index (SPI) SPI1 [(915 - 455) / (915 + 455)], SPI2 [(865 - 505) / (865 + 505)], and SPI3 [(915-495)/(915+495)]
- New Experimental Four Band (NEFB); NEFB1 [(780-660) / (780+660)], NEFB2 [(870-660)/ (870+660)], and NEFB3 [(970-660)/ (970+660)].

Table 5 f. Analysis of variance, single degree of freedom contrasts, treatment mean squares, and treatment means for forage yield, forage N and P content, spectral New-experimental 4 band sensor and, and digital picture indices at Feekes 10 growth stages, Perkins, OK, 2009

Growth stage	Source of variation	DF	Forage Mg ha^{-1}	Forage P g kg^{-1}	Forage N g kg^{-1}	NDVI	Spectral indices			New experimental 4-band indices			D Picture indices	
							SPI 1	SPI 2	SPI 3	NEFB 1	NEFB 2	NEFB 3	R/G	B/R
Feekes 10_2009	N rate	3	36.1***	.035***	.774***	Ns	.018*	.041**	.035***	Ns	Ns	Ns	Ns	Ns
	P rate	2	21.27***	.017***	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	NS
	N *P rate	6	5.5**	Ns	Ns	Ns	Ns	NS		Ns	Ns	NS	NS	Ns
	R-square	-	.88	.90	.68	Ns	.56	.70	.71	-	-	-	-	-
	Contrast													
	Linear N rate	**1**	77.7***	.035***	1.828	Ns	.0429 *	.081***	.083***	NS	NS	NS	NS	NS
	Quadratic N rate	1	30.6***	.066***	NS	NS	.044*	.04**	.021*	NS	NS	NS	NS	NS
	Linear Prate	1	41.8** *	.032***		NS	NS	NS	NS	NS	NS	NS	NS	NS
	Quadratic P rate	1	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
	Linear N*P rate	1	27.9***	.003***	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
	Treatment Means													
	N rate, kg ha-1	0	0.439	0.313	1.288	0.462	0.332	0.575	0.543	0.701	0.740	0.743	0.916	0.705
		56	3.645	0.177	1.174	0.566	0.425	0.672	0.626	0.604	0.660	0.670	0.872	0.605
		112	4.884	0.183	1.621	0.577	0.465	0.731	0.682	0.583	0.641	0.660	0.807	0.572
		168	4.405	0.219	1.810	0.549	0.42	0.696	0.668	0.560	0.630	0.650	0.824	0.65
		SED	0.50	0.08	0.236	0.161	0.13	0.11	0.105	0.186	0.169	0.16	0.189	0.167
	P rate, kg ha-1	0	1.3921	0.181	1.467	0.518	0.414	0.669	0.624	0.621	0.673	0.685	0.856	0.623
		34	3.55	0.235	1.529	0.496	0.436	0.690	0.646	0.583	0.642	0.660	0.848	0.637
		67	4.56	0.253	1.143	0.601	0.382	0.65	0.619	0.632	0.684	0.700	0.859	0.640
		SED	0.43	0.067	0.204	0.14	0.113	0.097	0.091	0.161	0.146	0.14	0.163	0.144

- NS,*, **, ***, not significant, significant at P < 0.05, P < 0.01, P < 0.001, respectively with their mean square values.
- SED – standard error of the difference for two equally replicated means, forage N or P = forage N or P content.
- Phosphorus Index (SPI) SPI1 [(915 - 455) / (915 + 455)], SPI2 [(865 - 505) / (865 + 505)], and SPI3 [(915-495)/(915+495)]
- New Experimental Four Band (NEFB); NEFB1 [(780-660) / (780+660)], NEFB2 [(870-660)/ (870+660)], and NEFB3 [(970-660)/ (970+660)].

Table 6. Analysis of variance, single degree of freedom contrasts, treatment mean squares and treatment means for grain yield, grain and soil N and P content, Perkins, OK, 2008 - 2009

				Year			2008				2009
	DF	**Grain yield**	**Soil P**	**Soil N**	**Grain P**	**Grain N**	**Soil P**	**Soil N**	**Grain P**	**Grain N**	**Grain yield**
		Mg kg^{-1}	--				g kg^{-1}	---			Mg kg^{-1}
N rate	3	9.03***	NS	NS	NS	76.86**	NS	NS	NS	64.74***	.51***
P rate	2	NS	.000***	NS	NS	NS	.014***	NS	1.2**	NS	NS
N *P rate	6	NS	NS	NS	NS	NS	.001*	NS		NS	NS
R-square	-	.73	.83	-	-	.55	.75	-	.55	.60	.58
Contrast											
Linear N rate	1	23.26***	NS	NS	NS	NS	NS	NS	NS	144***	.87***
Quadratic N rate	1	2.56*	NS	NS	NS	55.46*	NS	NS	NS	NS	.514**
Linear Prate	1	NS	.001***	NS	NS	NS	.028***	NS	1.61**	NS	NS
Quadratic P rate	1	NS	NS	NS	NS	NS	NS	NS	.78*	NS	NS
Linear N*P	1	NS	.000**	NS	NS	NS	NS	NS	NS	NS	NS
					Treatment Means						
N rate, kg ha-1	0	1.867	0.012			15.578	0.071		4.269	17.246	0.194
	56	2.783	0.019			16.367	0.069		4.270	18.593	0.683
	112	4.005	0.018			17.379	0.065		4.420	23.016	0.656
	168	3.856	0.021			22.097	0.069		4.490	21.733	0.667
	SED	0.394	0.03			0.88	0.07		0.296	0.732	0.229
P rate, kg ha-1	0	3.067	0.011			16.233	0.032		3.100	20.381	0.525
	34	3.067	0.020			18.600	0.073		4.571	20.600	0.579
	67	3.25	0.027			19.273	0.101		4.517	19.460	0.546
	SED	0.345	0.082			0.766	0.06		0.256	0.634	0.198

NS,*, **, ***, Not significant, significant at P< 0.05, P < 0.01, P < 0.001, respectively with their mean square values, GY-grain yield; grain N or P = grain N or P content

Table 7. Correlation analysis for the relationship between forage yields, forage N and P contents, and grain yield, grain N and P contents at three Feekes growth stages, Perkins, OK, 2008 – 2009

Growth stage		Year 2008			2009		
	source	Grain yield	Grain N	Grain P	Grain yield	Grain N	Grain P
		Mg kg^{-1}	------ g kg^{-1} -------		Mg kg^{-1}	---------- g kg^{-1}------------	
Feekes 4	Forage yield	.63***	.54***	NS	.54*	.61**	NS
	Forage P content	.35*	.47**	NS	NS	NS	NS
	Forage N content	NS	.NS	NS	.67**	.46*	NS
Feekes5/7	Forage yield	.63***	.54***	NS	.54*	.48*	.49*
	Forage P content	NS	.45**	.34*	.53*	.55*	NS
	Forage N content	.68***	.49**	.NS	.76***	.64***	NS
Feekes 10	Forage yield	.83***	.49**	NS	.58*	.71***	NS
	Forage P content	NS	NS	NS	.51*	.NS	NS
	Forage N content	.50**	.53***	NS	.NS	NS	NS

NS,*, **, ***, not significant, significant at P < 0.05, P < 0.01, P < 0.001, respectively with their correlation coefficient (r) values
Feekes5 for 2008 and Feekes 7 for 2009.

Table 8a. Stepwise regression analysis to select a candidate wavelength at P< 0.15 significance level to develop an index that can detect winter wheat forage N and P, grain N and P and forage and grain yield, at Feekes 5, 2008, Perkins, OK.

Feekes 5_2008			
No. of Variables	Wavelength/Index	Forage N (g kg^{-1})	
		R^2	CV
4	485, 655, 685, 695	0.64	-18.55
3	485, 685, 695	0.61	-19.9
2	485, 695	0.44	-18.3
	Index		
3	485/685, 485/695, 655/685	0.57	10.78
2	485/685, 485/695	0.45	20.3
1	485/685	0.29	32.7
		Forage P, (g kg^{-1})	
6	425,435 ,515, 585, 735, 745	0.78	-15.4
5	435 ,515, 585, 735, 745	0.74	-16.16
4	435 ,515, 585, 735, 895	0.70	-15.5
3	515, 585, 735, 895	0.65	-7.92
1	895	0.25	-7
		Grain N (g kg^{-1})	
8	435, 455, 535, 555, 635, 755, 785	0.85	114
4	535,635,775,915	0.63	296.8
3	635,775,915	0.51	398.1
2	775,915	0.45	444.3
1	775	0.17	683.2
	index		
3	535/775, 535/915, 635/915	0.51	3.02
2	535/775, 635/915	0.47	3.4
1	535/775	0.16	21.27
		Grain P, (g kg^{-1})	
2	955,975	0.16	115.5
1	975	0.07	127.7
		Grain yield, (Mg kg^{-1})	
3	745, 775, 785	0.62	-17.27
2	745, 785	0.6	-18.5
1	785	0.24	-10.3
		Forage yield , (Mg kg^{-1})	
7	515, 595, 605, 615, 665, 695, 935	0.86	149861.7
6	515, 595, 605, 665, 695, 935	0.84	172763
5	515, 595, 665, 695, 935	0.82	195262
4	515, 595, 695, 935	0.78	233660
3	515, 595, 935	0.76	260143
2	515, 935	0.71	310809.7
	index		
2	485/935, 615/695	0.67	0.48
1	485/695	0.59	5.65

- Indices were developed as a ration in the form of x/y.

Table 8b. Stepwise regression analysis to select a candidate wavelength at P< 0.15 significance level to develop an index that can detect winter wheat forage N and P, grain N, and forage and grain yield, at Feekes 10, 2008, Perkins, OK.

Feekes 10_2008			
No. of Variables	Wavelength/Index	Forage N (g kg^{-1})	
		R^2	CV
5	445, 465, 55, 675, 995	0.84	6.89
4	445, 555, 675, 995	0.82	8.9
3	465, 675, 995	0.70	11.77
2	555, 995	0.70	29.5
1	555	0.39	86.6
	Index		
5	445/675, 465/995, 555/675, 555/995	0.84	6.89
4	445/675, 465/995, 505/675, 555/995	0.62	31.5
2	505/675, 555/995	0.56	7.65
		Forage P, (g kg^{-1})	
16	405,445,455,475,495,535,555,575,575, 605, 715, 755,775, 795, 805, 815, 725	0.96	
1	475	0.2	57.14
		Grain N (g kg^{-1})	
3	455, 835, 845	0.47	
2	455, 835	0.33	3.00
1	455	0.27	3.86
		Grain yield, (Mg kg^{-1})	
5	645, 665, 675, 835, 985	0.84	
2	835, 985	0.75	
2	685, 835	0.67	2.92
1	665	0.60	7.78
	index		
1	465/835	0.65	0.19
		Forage yield, (Mg kg ha^{-1})	
4	435, 895, 915, 985	0.94	5
3	895, 915, 985	0.93	9.14
2	895, 985	0.93	18.6
1	895	0.73	105.6
	index		
3	435/895, 435/915, 435/985	0.93	4.0
2	435/895, 435/985	0.92	6.0
1	435/895	0.63	0126.47

- Indices were developed as a ration in the form of x/y.

Table 8c. Stepwise regression analysis to select a candidate wavelength at P< 0.15 significance level to develop an index that can detect winter wheat forage N and P, grain N and forage and grain yield, at Feekes 7, 2009, Perkins, OK.

Feekes 7_2009			
No. of Variables	Wavelength/Index	Forage N (g kg^{-1})	
		R^2	CV
4	485, 565, 585, 495	0.54	-0.92
3	485, 565, 585	0.51	-1.08
2	485, 565	0.47	-0.4
1	585	0.33	4.45
	index		
2	495/585, 565/865	0.55	0.42
1	495/585	0.52	0.64
		Forage P, (g kg^{-1})	
8	765, 485,625, 515, 645, 665, 475, 545	0.71	0.9
5	765, 485, 625, 515, 475	0.61	5.0
2	515, 475	0.16	3.0
	index		
2	685/765,475/765	0.31	3.9
		Grain N (g kg^{-1})	
2	995, 775	0.47	3
1	775	0.41	5.16
		Grain yield, (Mg kg^{-1})	
3	445, 565, 465	0.54	18.4
2	565, 645	0.47	23.0
1	645	0.43	25.5
		Forage yield, (Mg kg ha^{-1})	
6	485, 595, 665, 745, 765, 935	0.91	7
4	595, 665, 745, 765	0.88	12
3	665, 745, 765	0.84	22
2	665, 765	0.80	33
1	665	0.76	44.56
	index		
2	485/765, 665/745	0.82	13.3

- Indices were developed as a ration in the form of x/y.

Table 8d. Stepwise regression analysis to select a candidate wavelength at P< 0.15 significance level to develop an index that can predict mid season winter wheat forage N and P, grain N and P and forage and grain yield, at Feekes 10, 2009, Perkins, OK.

Feekes 10_2009			
No. of Variables	Wavelength/Index	Forage N (g kg^{-1})	
		R^2	CV
4	695, 845, 665, 535	0.68	-22.7
3	665, 695, 845	0.66	-24.5
2	695, 845	0.41	-24.0
1	695	0.28	-24.7
	index		
3	535/845, 535/665, 665/695	0.59	6
2	535/845, 665/695	0.56	16.4
1	535/695	0.4	31.5
		Forage P, (g kg^{-1})	
4	425, 545, 685, 775	0.6	-18.55
3	545, 685, 775	0.56	-19.8
2	545, 685	0.45	-19.8
1	685	0.27	-18.41
	index		
2	545/775, 545/685	0.58	2.24
1	545/685	0.42	12.6
		Grain N (g kg^{-1})	
5	485, 675, 685, 715, 755	0.67	3
4	485, 675, 685, 715	0.64	3.78
3	485, 675, 685	0.58	7
2	485, 685	0.47	013.9
1	685	0.35	21.8
	index		
4	715/755, 715/965, 485/965, 715/965	0.72	8.7
3	485/965, 715/755, 715/965	0.61	20.2
2	715/965	0.56	24.9
1	715/755	0.44	37.4
		Grain yield, (Mg kg^{-1})	
2	685, 475	0.41	29.83
1	685	0.36	32.85
		Forage yield, (Mg kg ha^{-1})	
2	685, 485	0.40	-15.1
1	685	0.35	-15.6

- Indices were developed as a ration in the form of x/y.

Table 9a. Selected separate wave lengths averaged from 10 nm band width that were significantly affected by N rate with the R^2 value greater than or equal to 0.65 and probability level ($P < 0.001$), and the contrast at Feekes 4 and 10, Perkins, OK, 2008 - 2009.

Growth stage	Average wave length at 10 nm band width	Model R^2 value	Model probability	Probability of the Contrast	
				Linear N rate	Quadratic N rate
Feekes 10,2008	465	.65	**	***	NS
	475	.65	**	***	NS
	505	.7	***	***	NS
	535	.67	**	***	NS
	565	.67	**	***	Ns
	575	.67	**	***	NS
	585	.66	**	***	NS
	595	.66	**	***	NS
	605	.69	***	***	NS
	615	.71	***	***	Ns
	625	.67	***	***	NS
	635	.7	***	***	NS
	645	.68	***	***	NS
	655	.7	***	***	NS
	665	.73	***	***	Ns
	675	.7	***	***	NS
	685	.72	***	***	NS
	695	.68	***	***	NS
Feekes 4,2009	565	.65	**	***	**
	575	.68	***	***	***
	585	.71	***	***	***
	595	.74	***	***	***
	605	.74	***	***	***
	615	.75	***	***	***
	625	.76	***	***	***
	635	.77	***	***	***
	645	.78	***	***	***
	655	.78	***	***	***
	665	.79	***	***	***
	675	.78	***	***	***
	685	.79	***	***	***
	695	.78	***	***	***

NS,*, **, ***, not significant, significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively

Table 9b. Selected separate wave lengths averaged from 10 nm band width that were significantly affected by N rate with the model R^2 value greater than or equal to 0.65 and probability level ($P < 0.001$), and the contrast at Feekes 7, Perkins, OK, 2009

Growth stage	Average wave length at 10 nm band width	Model R^2 value	Model probability	Probability of the Contrast	
				Linear N rate	Quadratic N rate
Feekes 7,2009	445	.76	***	***	**
	465	.66	***	***	**
	475	.68	***	***	**
	485	.74	***	***	**
	495	.72	***	***	***
	505	.72	***	***	**
	515	.71	***	***	**
	525	.67	**	***	NS
	555	.75	***	***	NS
	565	.69	***	***	**
	575	.74	***	***	**
	585	.76	***	***	**
	595	.77	***	***	**
	605	.78	***	***	**
	615	.78	***	***	**
	625	.79	***	***	**
	635	.79	***	***	**
	645	.8	***	***	***
	655	.8	***	***	***
	665	.8	***	***	***
	675	.8	***	***	NS
	685	.81	***	***	***
	695	.8	***	***	***
	705	.76	***	***	**
	745	.74	***	***	**
	755	.75	***	***	**
	765	.78	***	***	**
	775	.76	***	***	**
	785	.76	***	***	**
	795	.76	***	***	**
	805	.75	***	***	**
	815	.75	***	***	**
	825	.75	***	***	**
	835	.75	***	***	**
	845	.75	***	***	**
	855	.74	***	***	**
	865	.74	***	***	**
	875	.74	***	***	**
	885	.72	***	***	**
	895	.71	***	***	**
	905	.68	***	***	**
	915	.66	***	***	**

NS,*, **, ***, not significant, significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively

Table 9c. Selected separate wavelengths averaged from 10 nm band width that were significantly affected by N rate with the model R^2 value greater than or equal to 0.65 and probability level ($P < 0.001$), and the contrast at Feekes 10, Perkins, OK, 2009

Growth stage	Average wave length at 10 nm band width	Model R^2 value	Model probability	Probability of the Contrast	
				Linear N rate	Quadratic N rate
Feekes 10,2009	655	.8	***	***	***
	665	.8	***	***	***
	675	.8	***	***	NS
	685	.81	***	***	***
	695	.8	***	***	***

NS,*, **, ***, not significant, significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively

Figure 1.The effect of rate of N and P and their interactions on forage yield and forage P content at three growth stages, Perkins, Ok, 2008

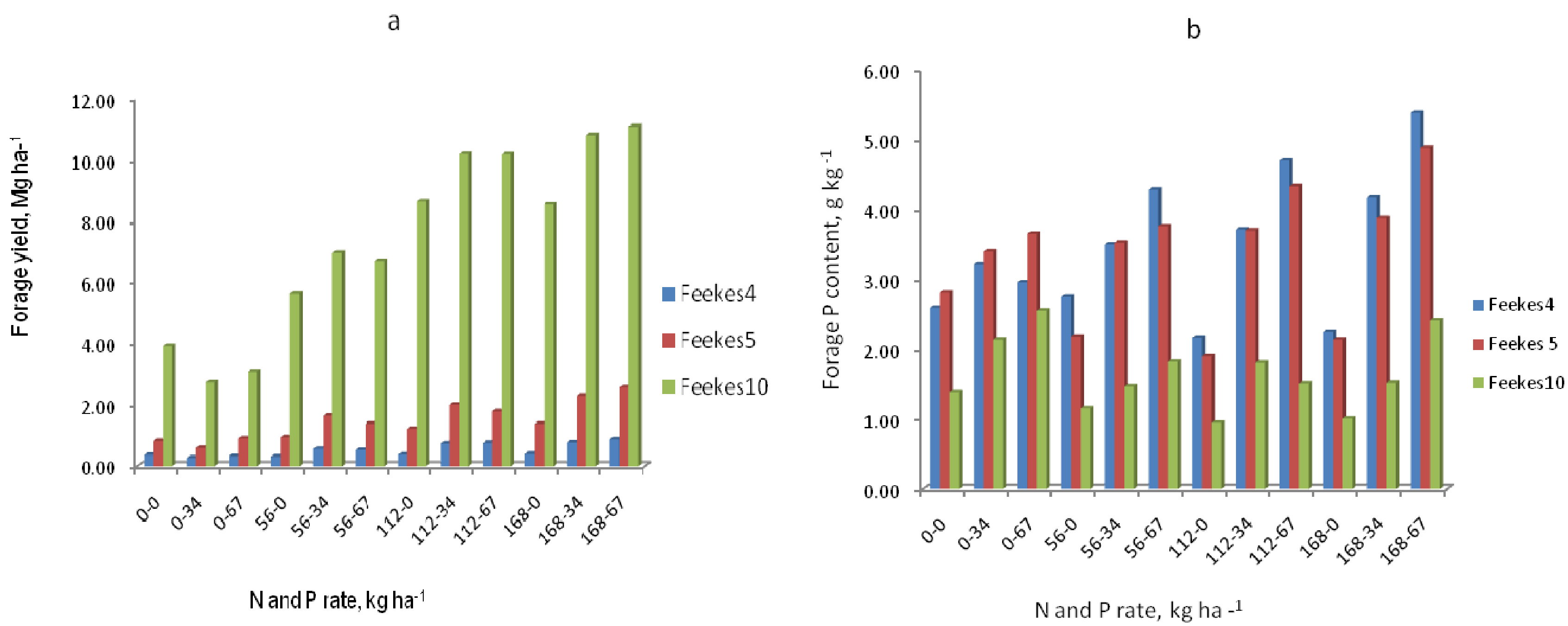


Figure 2.The effect of rate of N and P and their interactions on forage yield and forage P content at three growth stages, Perkins, OK, 2009

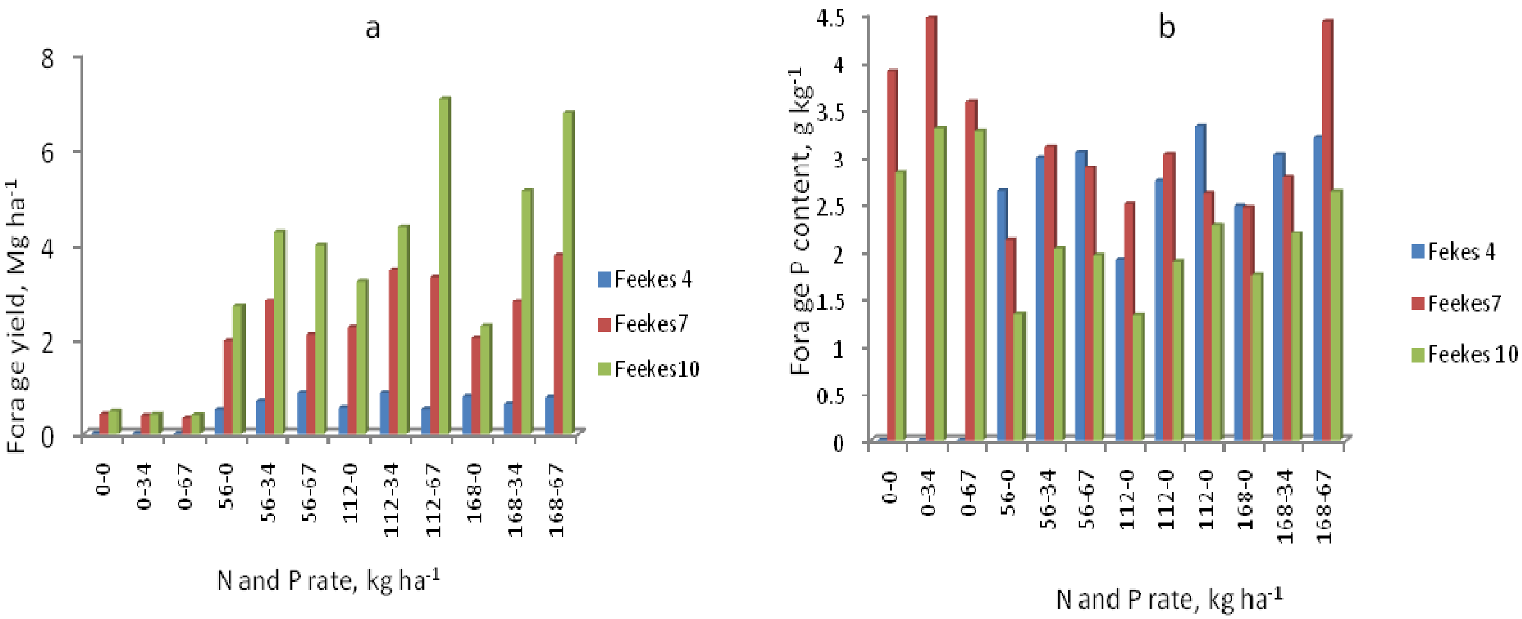
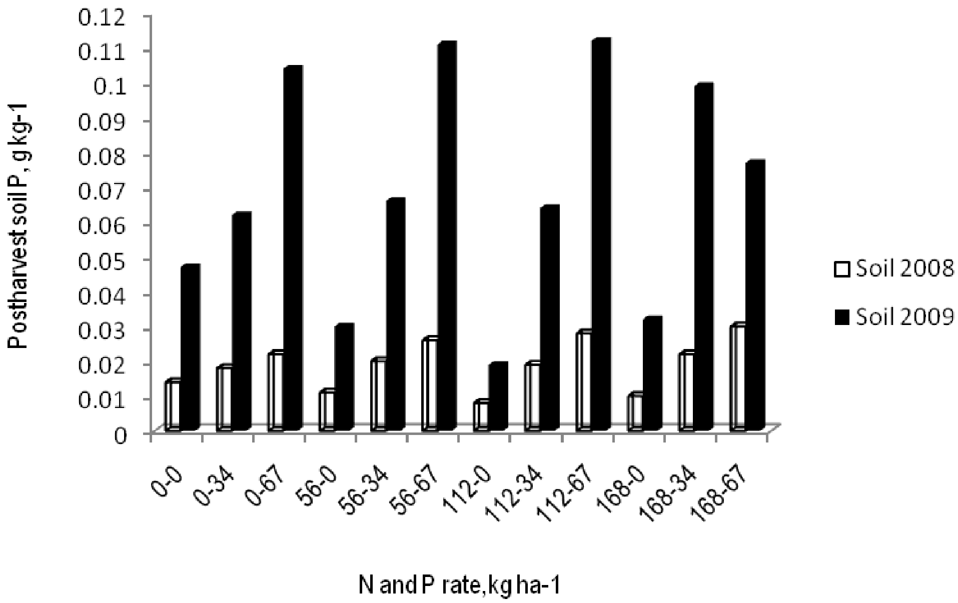


Figure 3.Comparison of postharvest residual soil P level at 15 cm depth by year and rate of fertilizer applied from least square means, Perkins, OK, 2008 - 2009



Appendices

Appendix 1a. Wavelengths at an average of 10 nm band width that were significantly ($P \leq 0.05$) correlated with forage N and P, and forage and grain yield at Feekes 4, 2008, Perkins, OK.

Feekes 4_2008				
Wavelength	Forage N, g kg^{-1}	Forage P, g kg^{-1}	Forage yield, Mg ha^{-1}	Grain yield, Mg ha^{-1}
405	NS	NS	NS	-0.34*
415	NS	NS	NS	-0.33*
425	NS	NS	NS	-0.34*
435	NS	NS	NS	-0.34*
445	NS	NS	NS	-0.36*
455	NS	NS	NS	-0.35*
465	NS	NS	NS	-0.34*
475	NS	NS	NS	-0.34*
485	NS	NS	NS	-0.36*
495	NS	NS	NS	-0.34*
505	NS	NS	NS	-0.34*
515	NS	NS	NS	-0.34*
525	NS	NS	NS	-0.33*
535	NS	NS	NS	-0.33*
545	NS	NS	NS	NS
555	NS	NS	NS	NS
565	NS	NS	NS	NS
575	NS	NS	NS	-0.35*
585	NS	NS	NS	-0.35*
595	NS	NS	NS	-0.34*
605	NS	NS	NS	-0.34*
615	NS	NS	NS	-0.34*
625	NS	NS	NS	-0.37
635	NS	NS	NS	-0.34*
645	NS	NS	NS	-0.33*
655	NS	NS	NS	NS
665	NS	NS	NS	-0.33
675	NS	NS	NS	-0.35*
685	NS	NS	NS	-0.35*
695	NS	NS	NS	-0.34*
705	NS	NS	NS	-0.33
715	NS	NS	NS	NS
725	NS	NS	NS	NS
735	NS	NS	NS	NS
745	NS	NS	NS	NS
755	NS	NS	NS	NS
765	NS	NS	NS	NS
775	NS	NS	NS	NS

- NS,*, **, ***, not significant, significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively
- Numbers with stars - correlation coefficient of the variable

Appendix 1b. Wavelengths at an average of 10 nm band width that were significantly ($P \leq 0.05$) correlated with forage N and P, forage and grain yield, Feekes 4, 2008, Perkins, OK.

Feekes 4_2008				
Wavelength	Forage N, g kg^{-1}	Forage P g kg^{-1}	Forage yield, Mg ha^{-1}	Grain yield, Mg ha^{-1}
785	NS	NS	NS	NS
795	NS	NS	NS	NS
805	NS	NS	NS	NS
815	NS	NS	NS	NS
825	NS	NS	NS	NS
835	NS	NS	NS	NS
845	NS	NS	NS	NS
855	NS	NS	NS	NS
865	NS	NS	NS	NS
876	NS	NS	NS	NS
885	NS	NS	NS	NS
895	NS	NS	NS	NS
905	NS	NS	NS	NS
915	NS	NS	NS	NS
925	NS	NS	NS	NS
935	NS	NS	NS	NS
945	NS	NS	NS	NS
955	NS	NS	NS	NS
985	NS	NS	NS	NS
965	NS	NS	NS	NS
975	NS	NS	NS	NS
985	NS	NS	NS	NS
995	NS	NS	NS	NS

- NS,*, **, ***, not significant, significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively
- Numbers with stars - correlation coefficient of the variable

Appendix 2a. Wavelengths at an average of 10 nm band width that were significantly ($P \leq 0.05$) correlated with forage N and P, forage and grain yield, Feekes 5, 2008, Perkins, OK.

Feekes 5_2008				
Wavelength	Forage N, g kg^{-1}	Forage P, g kg^{-1}	Forage yield, Mg ha^{-1}	Grain yield, Mg ha^{-1}
405	NS	NS	NS	NS
415	NS	NS	NS	NS
425	NS	NS	NS	NS
435	NS	NS	NS	NS
445	NS	NS	NS	NS
455	NS	NS	NS	NS
465	NS	NS	NS	NS
475	NS	NS	NS	NS
485	NS	NS	NS	NS
495	NS	NS	NS	NS
505	NS	NS	NS	NS
515	NS	NS	NS	NS
525	NS	NS	NS	NS
535	NS	NS	NS	NS
545	NS	NS	NS	NS
555	NS	NS	NS	NS
565	NS	NS	NS	NS
575	NS	NS	-0.34*	NS
585	NS	NS	-0.35*	NS
595	NS	NS	-0.36*	NS
605	NS	NS	-0.37*	NS
615	NS	NS	-0.39*	NS
625	NS	NS	-0.39*	NS
635	NS	NS	-0.40*	NS
645	NS	NS	-.041*	NS
655	NS	NS	-0.41*	NS
665	NS	NS	-0.41*	NS
675	NS	NS	-0.41*	NS
685	NS	NS	-0.42*	NS
695	NS	NS	-0.42*	NS
705	NS	NS	-0.38*	NS
715	NS	NS	NS	NS
725	NS	NS	NS	NS
735	NS	NS	NS	NS
745	NS	NS	NS	0.43**
755	NS	NS	NS	0.46**

- NS,*, **, ***, not significant, significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively
- Numbers with stars - correlation coefficient of the variable

Appendix 2b. wavelengths at an average of 10 nm band width that were significantly ($P \leq 0.05$) correlated with forage N and P, forage and grain yield, Feekes 5, 2008, Perkins, OK.

Feekes 5_2008				
Wavelength	Forage N, g kg^{-1}	Forage P g kg^{-1}	Forage yield, Mg ha^{-1}	Grain yield, Mg ha^{-1}
765	NS	NS	NS	0.48**
775	NS	NS	NS	0.48**
785	NS	NS	NS	0.48**
795	NS	NS	NS	0.49**
805	NS	NS	NS	0.48**
815	NS	NS	NS	0.47**
825	NS	NS	NS	0.47**
835	NS	NS	NS	0.46**
845	NS	NS	NS	0.46**
855	NS	NS	NS	0.46**
865	NS	NS	NS	0.45**
875	NS	NS	NS	0.46**
885	NS	NS	NS	0.44**
895	NS	NS	NS	0.43**
905	NS	NS	NS	0.41*
915	NS	NS	NS	0.39*
925	NS	NS	NS	0.37*
935	NS	NS	NS	0.35*
945	NS	NS	NS	NS
955	NS	NS	NS	NS
965	NS	NS	NS	NS
975	NS	NS	NS	NS
985	NS	NS	NS	NS
995	NS	NS	NS	NS
	NS	NS	NS	NS

- NS,*, **, ***, not significant, significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively
- Numbers with stars - correlation coefficient of the variable

Appendix 3a. Wavelengths at an average of 10 nm band width that were significantly ($P \leq 0.05$) correlated with forage N and P, forage and grain yield, Feekes 10, 2008, Perkins, OK.

Feekes 10_2008				
Wavelength	Forage N, g kg^{-1}	Forage P, g kg^{-1}	Forage yield, Mg ha^{-1}	Grain yield, Mg ha^{-1}
405	NS	NS	NS	NS
415	NS	NS	NS	NS
425	NS	NS	NS	NS
435	NS	NS	NS	NS
445	NS	0.38*	-0.42*	-0.42*
455	-0.40*	0.42*	-0.60***	-0.55**
465	-0.50*	0.46**	-0.74***	-0.67***
475	-0.53**	0.45**	-0.80***	-0.73***
485	-0.54**	0.47**	-0.81***	-0.73***
495	-0.53**	0.44**	-0.78***	-0.70***
505	-0.56**	0.45**	-0.81***	-0.72***
515	-0.57**	0.39*	-0.79***	-0.72***
525	-0.58**	0.39*	-0.78***	-0.71***
535	-0.60***	0.35*	-0.76***	-0.71***
545	-0.59***	0.35*	-0.68***	-0.63***
555	-0.63***	0.39*	-0.70***	-0.64***
565	-0.64***	0.36*	-0.69***	-0.69***
575	-0.63***	0.40*	-0.76***	-0.69***
585	-0..63	0.40*	-0.76***	-0.70***
595	-0.61	0.39*	-0.79***	-0.71***
605	-0.62***	0.41*	-0.80***	-0.72***
615	-0.61***	0.40*	-0.83***	-0.74***
625	-0.61***	0.40*	-0.83***	-0.74***
635	-0.61***	0.41*	-0.85***	-0.77***
645	-0.60***	0.42*	-0.85***	-0.74***
655	-0.60***	0.42*	-0.85***	-0.77***
665	-0.60***	0.41*	-0.86***	-0.77***
675	-0.58***	0.42*	-0.86***	0.76***
685	-0.59***	0.42*	-0.86***	-0.77***
695	-0.63***	0.40*	-0.82***	0-0.75***
705	-0.63***	NS	-0.64***	-0.58***
715	-0.59***	NS	-0.34*	NS
725	NS	NS	NS	NS
735	NS	NS	0.33*	NS
745	NS	NS	0.50**	0.40*
755	NS	NS	0.60**	0.50**

- NS,*, **, ***, not significant, significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively
- Numbers with stars - correlation coefficient of the variable

Appendix 3b. Wavelengths at an average of 10 nm band width that were significantly ($P \leq 0.05$) correlated with forage N and P, forage and grain yield, Feekes 10, 2008, Perkins, OK.

Feekes 10_2008				
Wavelength	Forage N, g kg^{-1}	Forage P g kg^{-1}	Forage yield, Mg ha^{-1}	Grain yield, Mg ha^{-1}
765	NS	NS	0.70***	0.60***
775	NS	NS	0.59***	0.50**
785	NS	NS	0.59**	0.50**
795	NS	NS	0.62***	0.54**
805	NS	NS	0.64***	0.56**
815	0.41*	NS	0.80***	0.69***
825	0.50**	NS	0.84***	0.72***
835	0.51**	NS	0.84***	0.71***
845	0.54**	NS	0.83***	0.70***
855	0.60***	NS	0.86***	0.71***
865	0.62***	NS	0.86***	0.71***
875	0.64***	NS	0.86***	0.71***
885	0.62***	NS	0.86***	0.69***
895	0.56***	NS	0.85***	0.70***
905	0.53**	NS	0.81***	0.68***
915	0.51**	NS	0.80***	0.67***
925	0.48**	NS	0.77***	0.64***
935	NS	NS	0.52**	0.41*
945	NS	NS	0.40**	NS
955	NS	NS	0.32*	NS
965	NS	NS	0.35*	NS
975	NS	NS	0.39*	NS
985	NS	NS	NS	NS
995	NS	NS	NS	NS

- NS,*, **, ***, not significant, significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively
- Numbers with stars - correlation coefficient of the variable

Appendix 4a. Wavelengths at an average of 10 nm band width that were significantly ($P \leq 0.05$) correlated with forage N and P, forage and grain yield, Feekes 7, 2009, Perkins, OK.

Feekes 7_2009				
Wavelength	Forage N, g kg^{-1}	Forage P, g kg^{-1}	Forage yield, Mg ha^{-1}	Grain yield, Mg ha^{-1}
405	-0.33*	NS	-0.58**	-0.50**
415	-0.37*	NS	-0.66***	-0.50**
425	-0.38*	NS	-0.68***	-0.55**
435	-0.43*	NS	-0.71***	-0.54**
445	-0.45*	NS	-0.77***	-0.63***
455	-0.49**	NS	-0.80***	-0.62***
465	-0.49**	NS	-0.80***	-0.62***
475	-0.49**	NS	-0.79***	-0.61***
485	-0.48**	NS	-0.81***	-0.62***
495	-0.50**	0.34*	-0.79***	-0.63***
505	-0.51**	0.31*	-0.82***	-0.63***
515	-0.56**	0.27*	-0.81***	-0.61***
525	-0.55**	0.28*	-0.77***	-0.60***
535	-0.53**	0.27*	-0.77***	-0.54**
545	-0.54**	0.22*	-0.77***	-0.56**
555	-0.55**	0.26*	-0.79***	-0.60***
565	-0.30**	0.30*	-0.80***	-0.58**
575	-0.57**	0.29*	-0.83***	-0.61***
585	-0.57**	0.32*	-0.84***	-0.63***
595	-0.56**	0.33*	-0.84***	-0.62***
605	-0.55**	0.33*	-0.85***	-0.62***
615	-0.57**	0.32*	-0.86***	-0.64***
625	-0.56**	0.35*	0.86***	-0.64***
635	-0.55**	0.35*	-0.87***	-0.64***
645	-0.55**	0.35*	-0.86***	-0.65***
655	-0.55**	0.35*	-0.86***	-0.64***
665	-0.55**	0.35*	-0.87***	-0.66***
675	-0.54**	0.36*	-0.87***	-0.65***
685	-0.54**	0.37*	-0.86***	-0.64***
695	-0.56**	0.34*	-0.86***	-0.62***
705	-0.58**	NS	-0.84***	-0.51**
715	-0.56**	NS	-0.72***	NS
725	NS	NS	NS	NS
735	43*	NS	0.73***	0.48**
745	0.51**	NS	0.81***	0.53**
755	0.53**	NS	0.82***	0.53**

- NS,*, **, ***, not significant, significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively
- Numbers with stars - correlation coefficient of the variable

Appendix 4b. Wavelengths at an average of 10 nm band width that were significantly ($P \leq 0.05$) correlated with forage N and P, forage and grain yield, Feekes 7, 2009, Perkins, OK.

Feekes 7_2009				
Wavelength	Forage N, g kg^{-1}	Forage P g kg^{-1}	Forage yield, Mg ha^{-1}	Grain yield, Mg ha^{-1}
765	0.55**	NS	0.85***	0.54**
775	0.54**	NS	0.84***	0.54**
785	0.53**	NS	0.83***	0.53**
795	0.53**	NS	0.84***	0.53**
805	0.54**	NS	0.82***	0.52**
815	0.53**	NS	0.82***	0.52**
825	0.53**	NS	0.81***	0.52**
835	0.52**	NS	0.82***	0.51**
845	0.52**	NS	0.82***	0.51**
855	0.52**	NS	0.82***	0.51**
865	0.52**	NS	0.81***	0.50**
875	0.52**	NS	0.81***	0.51**
885	0.51**	NS	0.80***	0.49**
895	0.51**	NS	0.79***	0.47**
905	0.49**	NS	0.76***	0.44**
915	0.48**	NS	0.74***	0.43*
925	0.46**	NS	0.70***	0.39*
935	NS	NS	0.46***	NS
945	NS	NS	NS	NS
955	NS	NS	NS	NS
965	NS	NS	NS	NS
975	NS	NS	NS	NS
985	NS	NS	NS	NS
995	NS	NS	NS	NS

- NS,*, **, ***, not significant, significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively
- Numbers with stars - correlation coefficient of the variable

Appendix 5a - wavelengths at an average of 10 nm band width that were significantly ($P \leq 0.05$) correlated with forage N and P, forage and grain yield, Feekes 10, 2009, Perkins, OK.

Feekes 10_2009				
Wavelength	Forage N, g kg^{-1}	Forage P g kg^{-1}	Forage yield, Mg ha^{-1}	Grain yield, Mg ha^{-1}
405	NS	NS	NS	NS
415	NS	NS	NS	NS
425	NS	NS	NS	NS
435	-0.34*	0.38*	-0.39*	-0.36*
445	-0.40*	0.41*	-0.45*	-0.48**
455	-0.40*	0.39*	-0.53**	-0.54**
465	-0.43**	0.42*	-0.52**	-0.53**
475	-0.43**	0.41*	-0.52**	-0.54***
485	-0.43**	0.41*	-0.52**	-0.53***
495	-0.42**	0.40*	-0.51**	-0.53***
505	-0.43**	0.40*	-0.50*	-0.52***
515	-0.43**	0.37*	-0.50**	-0.53***
525	-0.41**	NS	-0.48**	-0.51**
535	-0.40*	NS	-0.45**	-0.49**
545	-0.41*	NS	-0.43**	-0.46**
555	-0.42*	NS	-0.42**	-0.45**
565	-0.44**	NS	-0.43**	-0.45**
575	-0.47**	0.36*	-0.46**	-0.47**
585	-0.48**	0.39*	-0.49**	-0.50**
595	-0.49**	0.41*	-0.51**	-0.52***
605	-0.50**	0.41*	-0.52***	-0.53***
615	-0.50**	0.44**	-0.53***	-0.53***
625	-0.51**	0.45**	-0.54***	-0.55***
635	-0.51***	0.46**	-0.55***	-0.56***
645	-0.51***	0.48**	-0.56***	-0.57***
655	-0.52***	0.50**	-0.57***	-0.58***
665	-0.51**	0.51**	-0.58***	-0.59***
675	-0.52***	0.52**	-0.58***	-0.59***
685	-0.53***	0.52**	-0.59***	-0.60***
695	-0.49***	0.46**	-0.59***	-0.60***

- NS,*, **, ***, not significant, significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively
- Numbers with stars - correlation coefficient of the variable

Appendix 5b. Wavelengths at an average of 10 nm band width that were significantly ($P \leq 0.05$) correlated with forage N and P, forage and grain yield, Feekes 10, 2009, Perkins, OK.

Feekes 10_2009				
Wavelength	Forage N, g kg^{-1}	Forage P, g kg^{-1}	Forage yield, Mg ha^{-1}	Grain yield, Mg ha^{-1}
705	-0.30**	NS	-0.57***	-0.58**
715	-0.34*	NS	-0.46**	-0.47**
725	NS	NS	NS	NS
735	NS	NS	NS	NS
745	NS	-0.33*	NS	NS
755	0.35*	-0.34*	NS	NS
765	0.38*	-0.34*	NS	NS
775	0.36*	-0.34*	NS	NS
785	0.36*	-0.34*	NS	NS
795	0.37	NS	NS	NS
805	0.39*	NS	NS	NS
815	0.41*	NS	NS	NS
825	0.40*	NS	NS	NS
835	0.39*	NS	NS	NS
845	0.39*	NS	NS	NS
855	0.34*	NS	NS	NS
865	NS	NS	NS	NS
875	NS	NS	NS	NS
885	NS	NS	NS	NS
895	NS	NS	NS	NS
905	NS	NS	NS	NS
915	NS	NS	NS	NS
925	NS	NS	NS	NS
935	NS	NS	NS	NS
945	NS	NS	NS	NS
955	NS	NS	NS	NS
965	NS	NS	NS	NS
975	NS	NS	NS	NS
985	NS	NS	NS	NS
995	NS	NS	NS	NS

- NS,*, **, ***, not significant, significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively
- Numbers with stars - correlation coefficient of the variable

Appendix 6. Indices that were developed and tested to detect mid season winter wheat forage N and P status at Feekes 4 in 2008, Perkins, OK.

Feekes 4_2008						
Indices	Forage N	Forage P	Gain P	Grain N	Forage yield	Gain yield
985_405	NS	NS	NS	NS	NS	NS
915_415	NS	NS	NS	NS	NS	NS
845_415	NS	NS	NS	NS	0.36*	NS
915_455	NS	NS	NS	NS	0.45*	0.47**
865_455	NS	0.33*	NS	NS	0.50**	0.47**
755_475	NS	NS	NS	NS	0.47**	0.40*
815_465	NS	NS	NS	NS	0.51**	0.46**
865_505	NS	NS	NS	NS	0.45**	0.44**
725_515	NS	NS	NS	NS	0.44**	0.44**
915_505	NS	NS	NS	NS	0.44**	0.44**
705_505	NS	NS	NS	NS	0.35*	0.38*
675_555	NS	NS	NS	NS	NS	NS
865_555	NS	NS	NS	NS	0.46**	0.46**
915_555	NS	NS	NS	NS	0.36*	0.41*
785_585	NS	0.34*	NS	NS	0.49**	0.41*
745_615	NS	NS	NS	NS	0.49**	0.38*
785_665	NS	NS	NS	NS	0.44**	0.36*
805_705	NS	NS	NS	NS	0.45**	0.37*
755_645	NS	NS	NS	NS	0.43*	0.32*
915_495	NS	NS	NS	NS	0.40*	0.43**

- An index 985_405 means (985-405)/(985+405)

Appendix 7. Indices that were developed and tested to detect mid season winter wheat forage N and P status at Feekes 5in 2008, Perkins, OK.

Feekes 5_2008						
Indices	Forage N	Forage P	Gain P	Grain N	Forage yield	Gain yield
985_405	NS	NS	NS	NS	NS	NS
915_415	NS	NS	NS	0.50**	0.44**	0.55***
845_415	NS	NS	NS	0.53**	0.47**	0.57***
915_455	0.46**	0.50**	NS	0.44**	0.68***	0.47**
865_455	0.48**	0.49**	NS	0.54***	0.72***	0.57***
755_475	0.48**	0.47**	NS	0.46**	0.71***	0.53***
815_465	0.48**	0.48**	NS	0.49**	0.71***	0.55***
865_505	0.45**	0.44**	NS	NS	0.63***	0.37*
725_515	NS	0.39*	NS	NS	0.50**	NS
915_505	0.40*	0.39*	NS	NS	0.54***	NS
705_505	NS	NS	NS	-0.33*	-0.38*	-0.53***
675_555	-0.34*	-0.42**	NS	NS	-0.55***	-0.33*
865_555	0.49**	0.43**	NS	NS	0.64***	0.40*
915_555	0.43*	0.38*	NS	NS	0.55***	NS
785_585	0.50**	0.46**	NS	0.38*	0.68***	0.45**
745_615	0.47**	0.45**	NS	0.37*	0.66***	0.42**
785_665	0.47**	0.45**	NS	0.36*	0.66***	0.44**
805_705	0.54**	0.47**	NS	0.43**	0.73***	0.54***
755_645	0.48**	0.45**	NS	0.37*	0.67***	0.45**
915_495	0.40*	0.40*	NS	NS	0.55***	NS

- An index 985_405 means (985-405)/(985+405)

Appendix 8. Indices that were developed and tested to detect mid season winter wheat forage N and P status at Feekes 10 in 2008, Perkins, OK.

Feekes 10_2008						
Indices	Forage N	Forage P	Gain P	Grain N	Forage yield	Gain yield
985_405	NS	NS	NS	NS	NS	NS
915_415	0.37*	NS	NS	0.55***	0.60***	0.56***
845_415	0.51**	NS	NS	0.54***	0.80***	0.72***
915_455	0.55***	-0.35*	NS	0.57***	0.90***	0.76***
865_455	0.63***	-0.33*	NS	0.51**	0.91***	0.78***
755_475	0.46**	NS	NS	0.33*	0.85***	0.72***
815_465	0.51**	NS	NS	0.47**	0.93***	0.80***
865_505	0.63***	-0.36*	NS	0.45***	0.93***	0.79***
725_515	NS	NS	NS	NS	0.62***	0.55***
915_505	0.61***	-0.37*	NS	0.49***	0.93***	0.80***
705_505	-0.50**	NS	NS	NS	NS	NS
675_555	-0.49***	0.34*	NS	-0.34*	-0.87***	-0.74***
865_555	0.71***	-0.35*	NS	0.46***	0.91***	0.79***
915_555	0.69***	-0.33*	NS	0.50***	0.90***	0.79***
785_585	0.55***	-0.33*	NS	0.32*	0.88***	0.78***
745_615	0.52***	NS	NS	NS	0.87***	0.75***
785_665	0.64***	-0.35*	NS	0.34*	0.89***	0.78***
805_705	0.51***	-0.34*	NS	0.34*	0.91***	0.82***
755_645	0.60***	-0.34*	NS	0.46**	0.89***	0.77***
915_495	NS	-0.37	NS	NS	0.93***	0.81***

- An index 985_405 means (985-405)/(985+405)

Appendix 9. Indices that were developed and tested to detect mid season winter wheat forage N and P status at Feekes 4 in 2009, Perkins, OK.

Feekes 4_2009						
Indices	Forage N	Forage P	Gain P	Grain N	Forage yield	Gain yield
985_405	-	-	NS	0.38*	0.41*	NS
915_415	-	-	NS	0.43*	0.41*	NS
845_415	-	-	NS	0.48**	NS	NS
915_455	-	-	NS	0.44**	NS	NS
865_455	-	-	NS	0.46**	NS	NS
755_475	-	-	NS	0.49**	NS	0.37*
815_465	-	-	NS	0.48**	NS	NS
865_505	-	-	NS	0.42**	NS	0.40*
725_515	-	-	NS	0.47*	NS	NS
915_505	-	-	NS	NS	NS	0.37*
705_505	-	-	NS	-0.55**	NS	NS
675_555	-	-	NS	0.46**	NS	-0.60***
865_555	-	-	NS	0.53**	NS	0.44*
915_555	-	-	NS	0.50**	NS	0.42*
785_585	-	-	NS	0.49**	NS	0.50**
745_615	-	-	NS	0.57***	NS	0.52**
785_665	-	-	NS	0.59***	NS	0.53***
805_705	-	-	NS	0.59***	NS	0.55***
755_645	-	-	NS	0.63***	NS	0.52**
915_495	-	-	NS	0.45**	NS	0.36*

- An index 985_405 means (985-405)/(985+405)

Appendix 10. Indices that were developed and tested to detect mid season winter wheat forage N and P status at Feekes 7 in 2009, Perkins, OK.

Feekes 7_2009						
Indices	Forage N	Forage P	Gain P	Grain N	Forage yield	Gain yield
985_405	0.55***	NS	NS	0.72***	0.83***	0.56***
915_415	0.55***	NS	NS	0.71***	0.90***	0.61***
845_415	0.55***	NS	NS	0.70***	0.90***	0.61***
915_455	0.56***	NS	NS	0.69***	0.90***	0.63***
865_455	0.56***	NS	NS	0.69***	0.90***	0.62***
755_475	0.56***	NS	NS	0.68***	0.89***	0.64***
815_465	0.56***	NS	NS	0.68***	0.90***	0.63***
865_505	0.56***	-0.32*	NS	-0.68***	0.89***	0.63***
725_515	0.53***	-0.35*	NS	0.63***	0.87***	0.65***
915_505	0.56***	NS	NS	0.68***	0.89***	0.62***
705_505	NS	NS	NS	-0.34*	NS	NS
675_555	-0.51***	0.35*	NS	-0.60***	-0.86***	-0.62***
865_555	0.60***	NS	NS	0.70***	0.89***	0.61***
915_555	0.61***	NS	NS	0.71***	0.88***	0.59***
785_585	0.58***	NS	NS	0.69***	0.89***	0.63***
745_615	0.57***	NS	NS	0.68***	0.88***	0.63***
785_665	0.55***	-0.33*	NS	0.68**	0.88***	0.64***
805_705	0.59***	NS	NS	0.70***	0.88***	0.61***
755_645	0.56***	NS	NS	0.68***	0.88***	0.64***
915_495	0.57**	NS	NS	0.68***	0.88***	0.62***

- An index 985_405 means (985-405)/(985+405)

Appendix 11. Indices that were developed and tested to detect mid season winter wheat forage N and P status at Feekes 10 in 2009, Perkins, OK

Feekes 10_2009						
Indices	Forage N	Forage P	Gain P	Grain N	Forage yield	Gain yield
985_405	NS	NS	NS	NS	NS	NS
915_415	NS	NS	NS	NS	NS	NS
845_415	NS	NS	NS	NS	NS	NS
915_455	0.43*	-0.47**	NS	0.48***	0.37*	NS
865_455	NS	-0.38*	NS	0.36*	NS	NS
755_475	NS	-0.40*	NS	0.35*	NS	NS
815_465	NS	-0.39*	NS	0.37*	NS	NS
865_505	0.55**	-0.57***	NS	0.61***	0.52**	0.49*
725_515	NS	-0.46**	NS	NS	NS	NS
915_505	0.57***	-0.52**	NS	0.61***	0.62***	0.57***
705_505	NS	NS	NS	NS	NS	NS
675_555	-0.48**	0.67***	NS	-0.58***	-0.51	-0.52**
865_555	0.61***	-0.50*	NS	0.64***	0.61***	0.56***
915_555	0.54***	-0.37*	NS	0.54	0.58***	0.54***
785_585	0.54***	-0.57***	NS	0.60***	0.54***	0.54***
745_615	0.50**	-0.57***	NS	0.57***	0.48**	0.49***
785_665	0.51**	-0.59***	NS	0.59***	0.52**	0.52**
805_705	0.63***	-0.52**	NS	0.45***	0.58***	0.57***
755_645	0.52**	-0.59***	NS	0.59***	0.51**	0.49**
915_495	0.57***	-0.54***	NS	0.61***	0.59***	0.55**

- An index 985_405 means (985-405)/(985+405)

VITA

BIREHANE KASAYE DESTA

Candidate for the Degree of

Master of Science

Thesis: IDENTIFICATION OF SPECTRAL BANDS TO DETECT NITROGEN AND PHOSPHORUS DEFICIENCIES IN WINTER WHEAT

Major Field: Plant and Soil Sciences

Biographical:

Personal Data: Born at Bekoji, Ethiopia, on April 27, 1979

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Name: BIREHANE KASAYE DESTA Date of Degree: July, 2009

Institution: Oklahoma State University Location: Stillwater, Oklahoma

Title of Study: IDENTIFICATION OF SPECTRAL BANDS TO DETECT NITROGEN AND PHOSPHORUS DEFICIENCIES IN WINTER WHEAT

Pages in Study: 72 Candidate for the Degree of Master of Science

Major Field: Plant and Soil Sciences

Scope and Method of Study: The scope of the study was limited to searching for spectral wavelengths or indices that can detect mid season winter wheat N and P deficiencies and P independent of N deficiency. A randomized complete block design with three replications was employed. Treatments included twelve factorial combinations of three rates of P (0, 34 and 67 kg P ha^{-1}) and four rates of N (0, 56,112,168 kg N ha^{-1}). Full band Ocean optics spectrometer 4000 was used to collect the spectral readings at three different growth stages. The reflectance of wavelengths from 400 to 1000 nm was partitioned in to 60 wavelengths averaged from 10 nm band widths. Finally, each of the wavelengths was correlated with forage N and P content at each growth stages. Stepwise regression procedure was used to select the best wavelength or ratios of wavelengths that can detect forage N and P status. Analysis of variance was employed to test the effect of N and P rates on the spectral readings. For comparison, other devices: Greenseeker™ optical sensor, New-experimental 4 band sensor, and digital pictures were used.

Findings and Conclusions: No wavelength or index could detect winter wheat forage P content independent of forage N content using the above device and methods in the two-year study. This was likely because 1) wavelengths that detect forage P content were found within the range of the wavelengths that can detect forage N status and, 2) Nitrogen rate affected crop biomass and resulted in forage P content dilution as the crop grows.

ADVISER'S APPROVAL: Dr.William R. Raun